

=> d que stat 122

L14	25934 SEA FILE=HCAPLUS ABB=ON	(?PROTEIN? OR ?PEPTIDE?) (W) (?MOLECUL?
	OR ?POPUL? OR ?CELL?)	
L15	5686 SEA FILE=HCAPLUS ABB=ON	L14 AND (?ISOLAT? OR ?DETECT?)
L16	2254 SEA FILE=HCAPLUS ABB=ON	L15 AND (?APPARATUS? OR ?MECHANISM?)
	OR ?EQUIP? OR ?CONTAIN?)	
L17	979 SEA FILE=HCAPLUS ABB=ON	L16 AND (?PRODUCT? OR ?COMPOS? OR
	?METHOD? OR KIT?)	
L18	303 SEA FILE=HCAPLUS ABB=ON	L17 AND (?LYSIS? OR ?FILT?)
L19	167 SEA FILE=HCAPLUS ABB=ON	L18 AND ?CELL?
L20	4 SEA FILE=HCAPLUS ABB=ON	L19 AND ?PORE?
L21	42 SEA FILE=HCAPLUS ABB=ON	L19 AND ?CHROMATOGR?
L22	46 SEA FILE=HCAPLUS ABB=ON	L20 OR L21

=> d ibib abs 122 1-46

L22 .ANSWER 1 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 2003:678384 HCAPLUS
 DOCUMENT NUMBER: 139:192457
 TITLE: Methods for high throughput screening of genomic, cDNA or synthetic libraries for protein production in filamentous fungi
 INVENTOR(S): Emalfarb, Mark A.; Punt, Peter J.; Van Zeijl, Cornelis; Van Den Hondel, Cornelius USA
 PATENT ASSIGNEE(S):
 SOURCE: U.S. Pat. Appl. Publ., 48 pp., Cont.-in-part of Appl.
 No. PCT/US00/101999.
 CODEN: USXXCO
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 3
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2003162218	A1	20030828	US 2001-834434	20010413
WO 2001025468	A1	20010412	WO 2000-US10199	20000413
W: AE, AG, AL, AM, AT, AU, AZ, CR, CU, CZ, DE, DK, DM, HU, ID, IL, IN, IS, JP, KE, LU, LV, MA, MD, MG, MK, SD, SE, SG, SI, SK, TJ, ZA, ZW, AM, AZ, BY, KG, KZ, DE, DK, ES, FI, FR, GB, GR, CF, CG, CI, CM, GA, GN, MW, MX, NO, NZ, PL, PT, RO, RU, IE, IT, LU, MC, NL, PT, SE, BF, BJ, ML, MR, NE, SN, TD, TG				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, CF, CG, CI, CM, GA, GN, MW, MX, NO, NZ, PL, PT, RO, IE, IT, LU, MC, NL, PT, SE, BF, BJ, ML, MR, NE, SN, TD, TG				
WO 2001079558	A1	20011025	WO 2001-US12335	20010413
WO 2001079558	C1	20030227		
W: AE, AG, AL, AM, AT, AU, AZ, CO, CR, CU, CZ, DE, DK, DM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, LC, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
BR 2001005795	A	20020618	BR 2001-5795	20010413
EP 1272669	A1	20030108	EP 2001-927056	20010413
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,				

IE, SI, LT, LV, FI, RO, MK, CY, AL, TR
 JP 2003530843 T2 20031021 JP 2001-576942 20010413
 PRIORITY APPLN. INFO.: WO 2000-US10199 A2 20000413
 WO 1999-NL618 A 19991006
 WO 2001-US12335 W 20010413

AB The invention provides a **method** for the expression of exogenous DNA libraries in filamentous fungi. The fungi are capable of processing intron-containing eukaryotic genes, and also can carry out post-translational processing steps such as glycosylation and protein folding. The invention provides for the use of fungi with altered morphol., which permits high-throughput screening and directed mol. evolution of expressed proteins. The same transformed fungi may be used to produce larger quantities of protein for **isolation**, characterization, and application testing, and may be suitable for com. production of the protein as well.

L22 ANSWER 2 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 2003:591370 HCAPLUS
 DOCUMENT NUMBER: 139:130438
 TITLE: **Method for isolating** nucleic acids and protein from a single sample
 INVENTOR(S): Bosnes, Marie
 PATENT ASSIGNEE(S): Dynal Biotech Asa, Norway; Dzieglewska, Hanna
 SOURCE: PCT Int. Appl., 72 pp.
 CODEN: PIXXD2
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003062462	A2	20030731	WO 2003-GB156	20030116
W:	AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW, AM, AZ, BY			
RW:	GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, SE, SI, SK, TR, BF, BJ, CE, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG			

PRIORITY APPLN. INFO.: GB 2002-927 A 20020116
 GB 2002-27239 A 20021121

AB The present invention comprises a **method of isolating** nucleic acid and protein from the same sample with solid supports, wherein nucleic acid and protein components **contained** in the sample become bound to distinct solid supports. The invention also allows for **kits for isolating** nucleic acid and protein from the same sample and for use of the **method of isolating** nucleic acid and protein for the anal. and/or comparison of mRNA and/or protein expression and/or their correlation to genomic information.

L22 ANSWER 3 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 2003:242515 HCAPLUS
 DOCUMENT NUMBER: 138:283071
 TITLE: Proteome-wide analysis of protein interactions by high throughput mass spectrometry

INVENTOR(S): Bader, Gary; Climie, Shane; Durocher, Daniel; Figeys, Daniel; Gruhler, Albrecht; Heilbut, Adrian Mark; Ho, Yuen; Moore, Lynda A.; Moran, Michael; Muskat, Brenda; Tyers, Michael

PATENT ASSIGNEE(S): MDS Proteomics, Inc., Can.; Mount Sinai Hospital and Samuel Lunenfeld Research Institute

SOURCE: PCT Int. Appl., 133 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003025213	A2	20030327	WO 2002-CA1440	20020923
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
US 2003162221	A1	20030828	US 2002-252749	20020923
PRIORITY APPLN. INFO.:			US 2001-323930P	P 20010921
			US 2001-341213P	P 20011030
			US 2002-345286P	P 20020104

AB Methods and reagents for high throughput anal. of protein-protein interaction networks using high-throughput mass spectrometric protein complex identification (HMS-PCI) are described. The method is faster and less demanding of time than two-hybrid screening and it is feasible to identify directly protein complexes on a proteome-wide scale. The method uses proteins labeled with an affinity tag, such as an antigen, as baits to capture binding partners. Complexes are purified by means of the affinity label and the components rapidly characterized by mass spectrometry. Using 10% of predicted yeast proteins as baits, 3,617 protein interactions covering 25% of the yeast proteome were identified. Numerous protein complexes were identified, including many new interactions in various signaling pathways and in the DNA damage response. Comparison of the HMS-PCI data set with interactions reported in the literature revealed an average threefold higher success rate in detection of known complexes compared with large-scale two-hybrid studies. Given the high degree of connectivity observed in this study, even partial HMS-PCI coverage of complex proteomes, including that of humans, should allow comprehensive identification of cellular networks.

L22 ANSWER 4 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 2002:944510 HCPLUS

DOCUMENT NUMBER: 137:381979

TITLE: Method for separating protein mixtures

INVENTOR(S): Wozny, Wojciech; Cahill, Michael A.

PATENT ASSIGNEE(S): Proteosys AG, Germany

SOURCE: Eur. Pat. Appl., 8 pp.

CODEN: EPXXDW

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 1264839	A1	20021211	EP 2001-113909	20010607
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
WO 2002098904	A1	20021212	WO 2002-EP6208	20020606
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				

PRIORITY APPLN. INFO.: EP 2001-113909 A 20010607

AB A **method** for separating protein mixts. is provided. The inventive **method** is especially useful in characterizing and/or identifying low abundance proteins of interest from complex mixts. of protein, such as from human whole **cell** lysate protein mixts., in the order of one to several attomole of target protein. After labeling proteins of a protein mixture the mixture is separated and subsequently at least one population of labeled proteins (e.g. a protein spot from two dimensional gel electrophoresis) is **isolated** from the protein mixture (e. g. by two dimensional gel electrophoresis). This **isolated protein population** is mixed with a protein mixture containing an excess amount of the same protein in unlabeled form. After further biochem. fractionation of the resulting protein mixture, fractions containing the labeled target protein of interest are identified by tracking the label, and unlabeled protein identical to the labeled protein is characterized and/or identified. The success of **method** is dependent upon comigration of identical labeled and unlabeled proteins during the biochem. fractionation procedure employed. Preferably the labeling of the proteins is performed by radioactive labeling, especially by radioiodination. The **method** is applicable to any analyte mols., not just proteins.

REFERENCE COUNT: 2 THERE ARE 2 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L22 ANSWER 5 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 2002:928119 HCAPLUS

DOCUMENT NUMBER: 138:20436

TITLE: **Method of identifying cellular**

regulators of adeno-associated virus (aav)

INVENTOR(S): Weitzman, Matthew D.; Cathomen, Anton J.

PATENT ASSIGNEE(S): USA

SOURCE: U.S. Pat. Appl. Publ., 16 pp.

CODEN: USXXCO

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE

US 2002182595 A1 20021205 US 2002-135984 20020429
 PRIORITY APPLN. INFO.: US 2001-286951P P 20010427
 AB The present invention relates to a genetic screening assay to identify mols. that interact with a viral regulatory element. The viral regulatory element may be derived from an adeno associated virus (AAV), and may optionally contain at least one inverted terminal repeats (ITR) or one or more regions thereof. The construct containing the viral regulatory sequence is linked to a reporter gene so that the reporter gene will be expressed in the presence of proteins or other mols. that bind to the viral regulatory element. The assay is beneficial for analyzing mols. that bind to viral regulatory regions, and may be also useful as an assay kit to examine such interactions.

L22 ANSWER 6 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 2001:786936 HCPLUS

DOCUMENT NUMBER: 135:308838

TITLE: Method of preparing antigen from equine infectious anemia virus and kit for detecting antibody or antigen of equine infectious anemia virus

INVENTOR(S): Yurov, K. P.; Tokarik, E. F.; Galatyuk, A. E.; Samuilenko, A. Ya.; Lyul'kova, L. S.; Pestova, G. V.; Ulasov, V. I.

PATENT ASSIGNEE(S): Russia

SOURCE: Russ., No pp. given

CODEN: RUXXE7

DOCUMENT TYPE: Patent

LANGUAGE: Russian

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
RU 2146150	C1	20000310	RU 1999-119469	19990915
			RU 1999-119469	19990915

PRIORITY APPLN. INFO.: RU 1999-119469 19990915

AB The method involves the use of the novel strain of equine infectious anemia virus N 3-K-VNIITIBP-VIEV for antigen preparing. The strain is grown in horse embryo skin and/or muscle, or lung, and/or kidney cells up to accumulation of viruses at titer 105 ID 50/mL. Viruses are concentrated by ultrafiltration 10-20-fold, the lipid envelope is separated by treatment with cold ether and an end product peptide (mol. mass is 24-26 kDa) carrying group-specific antigen determinants is obtained. After the concentration stage viral material can be subjected to purification by high-speed centrifugation and ultracentrifugation addnl. After separation of the lipid envelope, the antigen obtained is purified by affinity chromatog. addnl. In this case the end product is immunolectrophoretically pure peptide. The obtained cultural antigen is used for kit-making, for identification of antigen of equine infectious anemia virus. Kits for identification of antibodies are designated for diagnosis of equine infectious anemia in RDP or IFA. Kit for identification of antigen is designated for titration of antigen of equine infectious anemia virus in the process of culturing. Kit for identification of antibodies in RDP has cultural antigen and specific precipitating serum. Kit for identification of antibodies in IFA has cultural antigen for immobilization on solid carrier, polystyrene plate and antispecies Ig class G (IgG) conjugated with horse radish peroxidase. Kit for identification of antigen has cultural antigen for immobilization on solid carrier, polystyrene plate and IgG to equine infectious anemia virus conjugated with horse radish peroxidase. Kits for IFA have

gelatin, substrate for peroxidase reaction carrying out, detergent and buffer solns. also. Kits can contain pos. and neg. controls also.

L22 ANSWER 7 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 2001:545723 HCAPLUS
 DOCUMENT NUMBER: 135:142230
 TITLE: High purity lipopeptides, **lipopeptide micelles** and processes for preparing same
 INVENTOR(S): Kelleher, Thomas J.; Lai, Jan-jie; Decourcey, Joseph P.; Lynch, Paul D.; Zenoni, Maurizio; Tagliani, Auro R.
 PATENT ASSIGNEE(S): Cubist Pharmaceuticals, Inc., USA
 SOURCE: PCT Int. Appl., 94 pp.
 CODEN: PIXXD2
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2001053330	A2	20010726	WO 2001-US1748	20010118
WO 2001053330	A3	20020418		
WO 2001053330	C2	20021017		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
BR 2001007731	A	20021001	BR 2001-7731	20010118
EP 1252179	A2	20021030	EP 2001-903121	20010118
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
JP 2003520807	T2	20030708	JP 2001-553802	20010118
NO 2002003476	A	20020920	NO 2002-3476	20020719
PRIORITY APPLN. INFO.:			US 2000-177170P	P 20000120
			US 2000-735191	A 20001128
			WO 2001-US1748	W 20010118

AB The invention discloses highly purified daptomycin and to pharmaceutical compns. comprising this compound. The invention discloses a **method** of purifying daptomycin comprising the sequential steps of anion exchange **chromatog.**, hydrophobic interaction **chromatog.** and anion exchange **chromatog.**. The invention also discloses a **method** of purifying daptomycin by modified buffer enhanced anion exchange **chromatog.**. An improved **method** for producing daptomycin by fermentation of Streptomyces roseosporus is described. The invention also discloses HPLC **methods** for anal. of daptomycin purity. **Methods** of using **lipopeptide micelles** for purifying lipopeptide antibiotics, such as daptomycin, and using them therapeutically are disclosed. Thus, daptomycin was produced in a fermentation culture of S. roseosporus and partially purified daptomycin (9.9 Kg) was purified by **microfiltration** from 5500 L of fermentation broth. The partially purified daptomycin was further purified and resulted in a bulk daptomycin preparation with a purity of 91%. The daptomycin preparation contained 14 impurities as determined by HPLC anal. The daptomycin

preparation was applied to a Poros P150 anion exchange resin (PE Biosystems) in Tris buffer pH 7.0 containing 6M urea and allowed to bind to the resin. The resin was washed with 3 column vols. of buffer prior to initiation of a NaCl gradient in the same buffer. Alternatively, the contaminants can be effectively removed from the column with a fixed salt level of 30 mM NaCl. The elution of purified daptomycin from the resin occurred at approx. 300 mM NaCl during a 0 to 1000 mM NaCl gradient. Daptomycin eluted from the column was greater than 99% pure as measured by the "first" HPLC method. The purified daptomycin contained only one detectable daptomycin contaminant. Anhydrodaptomycin and B-isomer were undetectable (<0.01% contamination). The level of the unidentified contaminant was 0.1-0.5%.

L22 ANSWER 8 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 2001:221448 HCPLUS

DOCUMENT NUMBER: 134:323087

TITLE: High-performance liquid chromatography

coupled on-line with electrospray ionization mass spectrometry for the simultaneous separation and identification of the Synechocystis PCC 6803 phycobilisome proteins

AUTHOR(S): Zolla, L.; Bianchetti, M.

CORPORATE SOURCE: Dipartimento di Scienze Ambientali, Universita della Tuscia, Viterbo, I-01100, Italy

SOURCE: Journal of Chromatography, A (2001), 912(2), 269-279
CODEN: JCRAEY; ISSN: 0021-9673

PUBLISHER: Elsevier Science B.V.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The complete resolution of the protein components of phycobilisome from cyanobacterium Synechocystis 6803, together with their detection and determination of mol. mass, has successfully been obtained by the combined use

of HPLC coupled online with electrospray ionization mass spectrometry. The method proposed consists of the isolation of the light-harvesting apparatus of cyanobacterium, by simply breaking cells in low-ionic-strength buffer, and subsequent injection of the total mixture of phycobilisomes into a C4 reversed-phase column. Identification of proteins was performed by SDS-PAGE (SDS-PAGE) of the samples collected from HPLC or by measuring the protein mol. mass coupling HPLC with mass spectrometry. The latter method allows the simultaneous separation of the phycobiliproteins, phycocyanin and allophycocyanin, from linker proteins and their identification, which due to their similar amino acid sequence and their similar hydrophobicity, might not be detected by denaturing SDS-PAGE. Under the exptl. conditions used, the pigment phycobilin is not removed from the polypeptide backbone, determining the hydrophobicity of the phycoproteins and hence their interaction with the reversed-phase column as well as in determining the protein-protein interaction into the phycobilisome

aggregation. Removal of the pigment, in fact, abolishes HPLC separation, emphasizing the essential role that the pigments play in maintaining the unusual tertiary structure of these proteins.

REFERENCE COUNT: 32 THERE ARE 32 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L22 ANSWER 9 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1998:806675 HCPLUS

DOCUMENT NUMBER: 130:66807

TITLE: Preparation of chemically modified polypeptides for

treatment of patients with reduced counts of granulocyte or blood platelet
INVENTOR(S): Yamasaki, Motoo; Suzawa, Toshiyuki; Kobayashi, Ken;
 Konishi, Noboru; Akinaga, Shiro; Maruyama, Kumiko
PATENT ASSIGNEE(S): Kyowa Hakko Kogyo Co., Ltd., Japan
SOURCE: PCT Int. Appl., 44 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9855500	A1	19981210	WO 1998-JP2504	19980605
W: AU, BG, BR, CA, CN, CZ, HU, IL, JP, KR, MX, NO, NZ, PL, RO, SG, SI, SK, UA, US, VN, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
AU 9875512	A1	19981221	AU 1998-75512	19980605
AU 744085	B2	20020214		
EP 921131	A1	19990609	EP 1998-923147	19980605
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
NZ 334068	A	20000728	NZ 1998-334068	19980605
US 2002028912	A1	20020307	US 1999-230733	19990203
US 6583267	B2	20030624		
NO 9900560	A	19990326	NO 1999-560	19990205
US 2003195339	A1	20031016	US 2003-365418	20030213
PRIORITY APPLN. INFO.:			JP 1997-149342	A 19970606
			WO 1998-JP2504	W 19980605
			US 1999-230733	A3 19990203

AB Claimed are chemical modified polypeptides, in particular having granulocyte colony stimulating factor activity, wherein at least one of the hydroxyl groups of a **polypeptide mol.** has been modified with polyalkylene glycols; a process for producing these polypeptides; a therapeutic **method** for treating patients with reduced counts of granulocyte or blood platelet by the use of these polypeptides; and therapeutic compns. **containing** these polypeptides. Thus, 205.5 mg monomethoxypolyethylene glycol propionic acid N-hydroxysuccinimide ester (M-SPA-20,000, Shearwater Polymer Corp.) was added to a 4.6 mg/mL solution of human granulocyte colony stimulating factor (hG-CSF) analog, i.e. [Thr1, Leu3, Tyr4, Arg5, Ser17]-Met-hG-CSF, in a phosphate buffer (pH 7.5) and stirred at 4° overnight to give polyethylene glycol-modified hG-CSF derivs. which were purified by a **chromatog.** Sephadex S-400 column to give two mono-, one di-, and two tri(polyethylene glycol) derivs. of hG-CSF. The linkage positions of polyethylene glycol in the polypeptide were investigated by peptide mapping using V8 protease digestion and HPLC separation and mass spectroscopy of the peptide fragments for these mono(polyethylene glycol) derivs. In two mono(polyethylene glycol) derivs. **isolated**, polyethylene glycol was linked to N-terminal Met and the hydroxy group of serine at 66 position, resp. Mono- and di(polyethylene glycol) derivs. showed the enhancement of proliferation of NFS60 **cells** equal to that of hG-CSF analog. The mono(polyethylene glycol) derivative linked to the Ser66 was 1.06-1.13 times more active than one linked to the terminal Met for enhancing the proliferation of NFS60 **cells** and was more stable in freezing-melting cycle test and more stable to thermolysis than the latter derivative

REFERENCE COUNT: 11 THERE ARE 11 CITED REFERENCES AVAILABLE FOR THIS

RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L22 ANSWER 10 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1997:734295 HCPLUS
 DOCUMENT NUMBER: 128:164531
 TITLE: Gas chromatographic analysis of resveratrol in plasma, lipoproteins and cells after in vitro incubations
 AUTHOR(S): Blache, D.; Rustan, I.; Durand, P.; Lesgards, G.; Loreau, N.
 CORPORATE SOURCE: 7, Boulevard Jeanne d'Arc, Faculte de Medecine, Laboratoire de Biochimie des Lipoproteines, Inserm, Universite de Bourgogne, Dijon, CJF 93-10, Fr.
 SOURCE: Journal of Chromatography, B: Biomedical Sciences and Applications (1997), 702(1 + 2), 103-110
 CODEN: JCBBEP; ISSN: 0378-4347
 PUBLISHER: Elsevier Science B.V.
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB Resveratrol is a trihydroxystilbene present in certain red wines. It may play a role in the inhibition of lipoprotein oxidation and platelet activity. We have developed the first **method** to measure resveratrol in animal and human samples and to study its incorporation in vitro. After adding epicoprostanol as an internal standard, samples are subjected to lipid extraction in the presence of antioxidant and under dim light to minimize both denaturation and isomerization of the trans-resveratrol to the cis-form. Exts. were purified by cold acetone precipitation and the resveratrol-containing acetone phase was evaporated under nitrogen. The resveratrol was analyzed as a trimethylsilyl derivative by capillary gas **chromatog**. which resolved the cis- and trans-resveratrol (6.6 and 12.9 min, resp.). Analyses of samples spiked with pure trans-resveratrol (0.1 to 10 µg) indicated that the **method** was specific and gave excellent linearity and recovery (96.8) with a high reproducibility (coefficient of variation: 3.3). The **detection** limit was about 50 ng/mL. Applications show that resveratrol was incorporated into blood **cells** and lipoproteins after in vitro incubations with plasma, lipoproteins and **cells**.

REFERENCE COUNT: 26 THERE ARE 26 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L22 ANSWER 11 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1996:195392 HCPLUS
 DOCUMENT NUMBER: 124:311483
 TITLE: Semipreparative chromatographic method to purify the normal cellular isoform of the prion protein in nondenatured form
 AUTHOR(S): Pergami, Paola; Jaffe, Howard; Safar, Jiri
 CORPORATE SOURCE: Lab. Central Nervous System Studies, Natl. Inst. Health, Bethesda, MD, 20892, USA
 SOURCE: Analytical Biochemistry (1996), 236(1), 63-73
 CODEN: ANBCA2; ISSN: 0003-2697
 PUBLISHER: Academic
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB A fundamental step in the pathogenesis of spongiform encephalopathies (prion diseases) is the conversion of the **cellular** isoform of prion protein (PrPC) into the infectious form (scrapie isoform, PrPSc), apparently by a conformational **mechanism**. Comparison of the native secondary and tertiary structures of both proteins is essential to elucidate the mol. basis of this transformation. To obtain sufficient

quantities of native-like PrPC, we have developed a semipreparative method to purify PrPC from hamster brains. PrPC was solubilized from purified synaptosomal and microsomal membranes by the nonionic detergent n-octyl- β -glucopyranoside; the soluble fraction was loaded at pH 7.5 onto a semipreparative cation-exchange TSK-SP-5PW (HPLC) column. The fractions eluted by linear NaCl gradient and enriched for PrPC were sequentially purified using an immobilized ion-affinity HPLC column charged by Co²⁺, followed by wheat germ agglutinin (WGA)-affinity HPLC or size-exclusion HPLC (SE-HPLC) using a TSK G3000SW column. More than 95% purity was achieved after SE-HPLC as estimated by quant. densitometry of the silver-stained SDS-PAGE gel; the recovery of total brain PrPC was $\geq 8\%$. The purified PrPC was a monomer with an intact N-terminus, and with a Stoke's radius of 26 Å, corresponding to that expected from the mol. weight for a native protein. The presence of the native-like conformation was further verified by peptide mapping after limited trypsin proteolysis, and by the apparent unfolding in guanidine hydrochloride, as detected by SE-HPLC.

L22 ANSWER 12 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1995:949691 HCAPLUS

DOCUMENT NUMBER: 124:134351

TITLE: Derivatization of dipeptides with 4-fluoro-7-nitro-2,1,3-benzoxadiazole for laser-induced fluorescence and separation by micellar electrokinetic chromatography

AUTHOR(S): Beijersten, Ingegerd; Westerlund, Douglas

CORPORATE SOURCE: Department of Analytical Pharmaceutical Chemistry, Uppsala University Biomedical Centre, P.O. Box 574, S-751 23, Uppsala, Swed.

SOURCE: Journal of Chromatography, A (1995), 716(1 + 2), 389-99

CODEN: JCRAEY; ISSN: 0021-9673

PUBLISHER: Elsevier

DOCUMENT TYPE: Journal

LANGUAGE: English

AB In capillary electrophoresis generally very small sample vols. are introduced, which often gives problems regarding detns. of low concns. of the analytes. Frequently, therefore, they have to be transformed into products by suitable derivatization reagents. In this study 4-fluoro-7-nitro-2,1,3-benzoxadiazole (NBD-F) was used as a pre-capillary fluorogenic reagent for dipeptides used as model compds. to study the characteristics of the derivatization procedure. Main emphasis was put on optimization of the reaction conditions using a chemometric approach involving a fractional factorial design for screening expts. and a central composite face-centered design for response surface modeling. The reagent excess must be at least 70 times to get a linear response, the reaction mixture should consist of a phosphate buffer with low ionic strength (0.001) at pH 7 containing 15% of iso-PrOH. The presence of the micellar agent Brij 35 in the background electrolyte increased the fluorescence intensity of the analyte product at least 3 times, and the separation selectivity increased compared to using a neat buffer. Leu-Val, chosen as a model peptide for studies on quant. detns., could be determined at the level 10⁻⁷ M (2 fmol injected) with a quant. recovery and a relative standard deviation of 2.4%. The limit of detection was 4 + 10⁻⁹ M (70 amol injected).

L22 ANSWER 13 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1995:868553 HCAPLUS

DOCUMENT NUMBER: 123:250451

TITLE: Use of green fluorescent protein for visualization of

AUTHOR(S): Webb, Chris D.; Decatur, Amy; Teleman, Aurelio; Losick, Richard

CORPORATE SOURCE: Dep. Mol. Cell. Biol., Harvard Univ., Cambridge, MA, 02138, USA

SOURCE: Journal of Bacteriology (1995), 177(20), 5906-11
CODEN: JOBAAY; ISSN: 0021-9193

PUBLISHER: American Society for Microbiology

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The authors report the use of the green fluorescent protein (GFP) of *Aequorea victoria*. Sporangia bearing the gene (*gfp*) for the green fluorescent protein fused to genes under the control of the sporulation transcription factor σF exhibited a **forespore**-specific pattern of fluorescence. **Forespore**-specific fluorescence could be **detected** with fusions to promoters that are utilized with low (*csfB*) and high (*sspE-2G*) efficiency by σF - containing RNA polymerase. Conversely, a mother **cell**-specific pattern of fluorescence was observed in sporangia bearing a transcriptional fusion of *gfp* to a **spore** coat protein gene (*cotE*) under the control of σE and an in-frame fusion to a regulatory gene (*gerE*) under the control of σK . An in-frame fusion of *gfp* to *cotE* demonstrated that GFP can also be used to visualize **protein subcellular** localization. In sporangia producing the *CotE-GFP* fusion protein, fluorescence was found to localize around the developing **spore**, and this localization was dependent upon *SpoIVA*, a morphogenetic protein known to determine proper localization of *CotE*.

L22 ANSWER 14 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 1994:503633 HCPLUS
DOCUMENT NUMBER: 121:103633
TITLE: Complex combinatorial chemical libraries encoded with tags
INVENTOR(S): Still, W. Clark; OHL-Meyer, Michael H. J.; Wigler, Michael; Dillard, Lawrence; Reader, John
PATENT ASSIGNEE(S): Columbia University, USA; Cold Spring Harbor Lab.
SOURCE: PCT Int. Appl., 147 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 4
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9408051	A1	19940414	WO 1993-US9345	19931001
W: AT, AU, BG, BR, CA, CH, CZ, DE, DK, ES, FI, GB, HU, JP, KR, LU, NL, NO, NZ, PL, RO, RU, SE, US				
RW: AT, BE, CH, DE, DK, ES, FR, GB, IT, LU, NL, PT, SE				
EP 665897	A1	19950809	EP 1994-900350	19931001
EP 665897	B1	20030709		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE				
HU 72495	A2	19960528	HU 1995-952	19931001
JP 08506175	T2	19960702	JP 1993-509330	19931001
AU 686579	B2	19980212	AU 1994-55369	19931001
AU 9455369	A1	19940426		
IL 107166	A1	20001031	IL 1993-107166	19931001
AT 244769	E	20030715	AT 1994-900350	19931001

NO 9501230	A	19950330	NO 1995-1230	19950330
US 6001579	A	19991214	US 1995-485018	19950607
AU 9745258	A1	19980212	AU 1997-45258	19971117
AU 716621	B2	20000302		
PRIORITY APPLN. INFO.:			US 1992-955371	A 19921001
			US 1993-13948	A 19930204
			US 1993-130271	B2 19931001
			WO 1993-US9345	W 19931001
			US 1993-159861	B2 19931130
			US 1994-227007	A3 19940413

OTHER SOURCE(S): MARPAT 121:103633

AB **Methods** and compns. are provided for encoded combinatorial chemical, whereby at each stage of the synthesis, a support such as a particle upon which a compound is being synthesized is uniquely tagged to define a particular event, usually chemical, associated with the synthesis of the compound on the support. The tagging is accomplished using identifier mols. which record the sequential events to which the supporting particle is exposed during synthesis, thus providing a reaction history for the compound produced on the support. Various **products** can be produced in the multi-stage synthesis, such as oligomers and synthetic nonrepetitive organic mols. Conveniently, nested families of compds. can be employed as identifiers, where number and/or position of a substituent define the choice. Alternatively, **detectable** functionalities may be employed, such as radioisotopes, fluorescers, halogens, and the like, where presence and ratios of two different groups can be used to define stage or choice. Particularly, pluralities of identifiers may be used to provide a binary or higher code, so as to define a plurality of choices with only a few detachable tags. The particles may be screened for a characteristic of interest, particularly binding affinity, where the **products** may be detached from the particle or retained on the particle. The reaction history of the particles which are pos. for the characteristic can be determined by the release of the tags and anal. to define the reaction history of the particle. An encoded combinatorial library of 2401 peptides was prepared (by solid phase synthesis) having the sequence (X4)EEDLGGGG (X = Asp, Glu, Ile, Lys, Leu, Gln, or Ser). The 4 Gly served as a spacer between the encoded amino acid sequence and the bead. The library included the sequence KLISEEDL, part of the epitope bound by monoclonal antibody 9E10 to the human C-myc gene **product**. The identifiers used were 2-nitro-4-carboxybenzyl O-aryl-substituted ω -hydroxyalkyl carbonates (aryl = pentachlorophenyl, 2,4,6-trichlorophenyl, or 2,6-dichloro-4-fluorophenyl) and were attached via their carboxylic acids to tag free amino groups on each bead. The tags were released from each selected bead by **photolysis**, silylated, and analyzed by electron capture gas **chromatog**. The binary synthesis code of the bead was directly determined from the **chromatogram** of the tags.

L22 ANSWER 15 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1994:239988 HCPLUS

DOCUMENT NUMBER: 120:239988

TITLE: Virion-associated trans-regulatory protein of human T-cell leukemia virus type I

AUTHOR(S): Lillehoj, Erik P.; Alexander, Steve S.

CORPORATE SOURCE: Cambridge Biotech Corp., Rockville, MD, 20850, USA

SOURCE: AIDS Research and Human Retroviruses (1992), 8(2), 237-44

CODEN: ARHRE7; ISSN: 0889-2229

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Western blot anal. of HTLV-I virus particles from HUT-102 **cells**

revealed a 40-kD protein strongly reactive with Tax-specific rabbit antisera. This protein subsequently was **isolated** from d. gradient purified virions by preparative SDS-PAGE, purified from comigrating Gag and human **cellular** proteins by reversed-phase high-performance liquid **chromatog.** (HPLC) and identified as the tax-encoded gene **product** by amino acid **composition anal.** Among **extracellular** virions from 5 HTLV-I producing **cell** lines, only those from HUT-102 and C10MJ **cells contained** a **detectable** Tax protein, although all **cells** expressed Tax mRNA and **protein intracellularly.** To investigate the diagnostic implications of virion-associated Tax protein, sera from HTLV-I-infected individuals were compared on HUT-102 and MT-2 virus Western blots. The seroprevalence of antibodies to Tax, but not Gag or Env proteins, was substantially higher among adult T-**cell** leukemia and tropical spastic paraparesis patients using HUT-102 viral proteins. Thus, immunoassays utilizing HUT-102 virus are most sensitive for **detection** of Tax-reactive antibodies.

L22 ANSWER 16 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1993:512929 HCAPLUS

DOCUMENT NUMBER: 119:112929

TITLE: **Method and apparatus for detecting trace contaminants**

INVENTOR(S): Afeyan, Noubar B.

PATENT ASSIGNEE(S): Perseptive Biosystems, Inc., USA

SOURCE: PCT Int. Appl., 29 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9300584	A1	19930107	WO 1992-US5043	19920615
W: AU, CA, JP RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LU, MC, NL, SE				
CA 2111695	AA	19930107	CA 1992-2111695	19920615
AU 9222388	A1	19930125	AU 1992-22388	19920615
AU 660480	B2	19950629		
EP 591407	A1	19940413	EP 1992-914471	19920615
EP 591407	B1	19970416		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, MC, NL, SE				
JP 06510600	T2	19941124	JP 1993-501529	19920615
JP 3431142	B2	20030728		
AT 151880	E	19970515	AT 1992-914471	19920615
US 5306426	A	19940426	US 1993-67418	19930525
PRIORITY APPLN. INFO.:			US 1991-721192 A	19910626
			WO 1992-US5043 A	19920615

AB Trace contaminants (e.g. pyrogens) are **detected** in a solution comprising a major amount of a dissolved **product** (e.g. protein) by flowing the solution through means for extracting the **product** to produce an effluent flow free of the **product** (e.g. with a **product-specific affinity chromatog.** matrix); flowing the effluent through a trace solute-adsorbing means to accumulate the trace solute (e.g. with a protein-binding matrix); and eluting the trace solute from the adsorbent to produce an eluant fraction **containing** a **detectable** quantity of the trace solute. A schematic representation of 1 embodiment of the **apparatus** is shown.

L22 ANSWER 17 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1993:447798 HCAPLUS
 DOCUMENT NUMBER: 119:47798
 TITLE: Preparation of sample solution for determination of nitrate and nitrite in various foods by colorimetry and ion chromatography
 AUTHOR(S): Tsuji, Sumiko; Shibata, Tadashi; Ezaki, Masumi; Ito, Katuhiko; Sase, Katutoshi; Ito, Yoshio
 CORPORATE SOURCE: Osaka Branch, Natl. Inst. Hyg. Sci., Osaka, 540, Japan
 SOURCE: Shokuhin Eiseigaku Zasshi (1993), 34(2), 161-7
 CODEN: SKEZAP; ISSN: 0015-6426
 DOCUMENT TYPE: Journal
 LANGUAGE: Japanese
 AB A method for preparation of sample solution for determination of nitrate and nitrite in vegetables, beef, and their processed foods by colorimetry and suppressed ion chromatog. (IC) with UV detection was developed. Nitrate and nitrite were simultaneously extracted from foods with Na₂B4O₇ solution at 80°, then the exts. were cooled and filtered through an ultrafilter (Molcut II). Nitrite and nitrate were determined in the filtrate by colorimetry and IC, resp. In the case of determination of nitrate in foods containing >3% NaCl, the salt was eliminated from the exts. by an On Guard-Ag cartridge. The recoveries of nitrate were 90.0-102.0% from foods fortified at 5-1000 µg/g and those of nitrite were 84.2-101.0% from foods fortified at 5-50 µg/g.

L22 ANSWER 18 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1992:507519 HCAPLUS
 DOCUMENT NUMBER: 117:107519
 TITLE: Comparison of pretreatment methods for the determination of copper and iron in serum by HPLC with electrochemical detection
 AUTHOR(S): Nagaosa, Yukio; Ishida, Kazuhisa
 CORPORATE SOURCE: Fac. Eng., Fukui Univ., Fukui, 910, Japan
 SOURCE: Bunseki Kagaku (1992), 41(5), T73-T76
 CODEN: BNSKAK; ISSN: 0525-1931
 DOCUMENT TYPE: Journal
 LANGUAGE: Japanese.
 AB Three different deproteinization methods for the liquid chromatog. determination of Cu(II) and Fe(III) in sera have been investigated and compared. Samples containing Cu(II) and Fe(III) were treated with 8-hydroxyquinoline to form metal complexes, which were applied to octadecylsily (ODS) column and determined electrochem. The anal. results obtained with both trichloroacetic acid-HCl and MeOH-HCl are in good agreement with the recommended values. The method using perchloric acid gave neg. errors for the determination of Fe(III) in normal serum. For accurate determination the 2 metal complexes with 8-hydroxyquinoline should be formed prior to injection onto an ODS column.

L22 ANSWER 19 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1991:203109 HCAPLUS
 DOCUMENT NUMBER: 114:203109
 TITLE: Optic biosensor for use in organic solvents
 INVENTOR(S): Spohn, Uwe; Miethe, Peter; Voss, Harald
 PATENT ASSIGNEE(S): Martin-Luther-Universitaet Halle-Wittenberg, Ger. Dem. Rep.
 SOURCE: Ger. (East), 4 pp.
 CODEN: GEXXA8

DOCUMENT TYPE: Patent
 LANGUAGE: German
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
DD 278860	A1	19900516	DD 1988-324046	19881227
			DD 1988-324046	19881227

PRIORITY APPLN. INFO.: AB The title biosensor consists of a permeable support system (e.g. a polymer membrane or paper), a fiber-optic **detection** system, and a thin catalyst layer comprising a biocatalyst (enzyme, enzyme-labeled **protein, cells, organelles, etc.**) in a lyotropic mesophase which is insol. in, and chemical and phys. stable towards, the organic solvent. The mesophase consists of a ternary or pseudoternary surfactant/organic solvent/water system, where the organic solvent is immiscible with water. Thus, a biosensor comprised (1) 2 optic fibers in optical contact with (2) a quartz disk covered by (3) a lyotropic mesophase 0.1-1 mm thick **composed** of heptaoxyethylene tetradecyl ether 8.56, n-hexane 76.84, lactate dehydrogenase 0.05 weight%, and water, and (4) a double membrane comprising (a) a porous PTFE membrane with mean **pore** size 10-20 µm and (b) a perforated PTFE membrane in contact with the organic solvent **containing** the analyte (pyruvate), NADH, and a solubilizer (cetyltrimethylammonium bromide). The fluorescence of the NAD formed was measured with excitation at 340 nm and emission at 460 nm. Pyruvate was determined over the range 0.05-0.5 mM.

L22 ANSWER 20 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1989:589872 HCPLUS
 DOCUMENT NUMBER: 111:189872
 TITLE: Structural assessment of the N-linked oligosaccharides of **cell-CAM 105** by lectin-agarose affinity chromatography
 AUTHOR(S): Bierhuizen, Marti F. A.; Hansson, Magnus; Odin, Per;
 Debray, Henri; Oebrink, Bjoern; Van Dijk, Willem
 CORPORATE SOURCE: Fac. Med., Vrije Univ., Amsterdam, NL-1007 MC, Neth.
 SOURCE: Glycoconjugate Journal (1989), 6(2), 195-208
 CODEN: GLJOEW; ISSN: 0282-0080
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB The N-linked oligosaccharides of **cell-CAM 105**, a glycoprotein involved in the **intercellular** adhesion between rat hepatocytes, were studied by sequential lectin-agarose affinity **chromatog.** of desialylated, ¹⁴C-labeled glycopeptides. These glycopeptides were obtained by extensive Pronase digestion followed by N-[¹⁴C]acetylation of the peptide moieties and desialylation by mild acid **hydrolysis**. Assuming that all glycopeptides were radiolabeled to the same specific radioactivity, Con A-Sepharose **chromatog.** indicated that the majority of the glycans (84%) were of the complex-type of which .apprx.50% were biantennary structures. The remainder of the glycans comprised oligomannose-type structures and/or incomplete biantennary structures. Pea lectin-agarose **chromatog.** revealed that part of the biantennary glycans **contained** a fucose residue α (1-6)-linked to the N-acetylglucosamine which was attached to asparagine. Furthermore, the presence of tri- and tetra- and/or tri'-antennary complex-type glycans was demonstrated by **chromatog.** on immobilized Phaseolus vulgaris leukoagglutinating phytohemagglutinin and Aleuria aurantia lectin (AAL). AAL-agarose **chromatog.**

furthermore indicated the presence of $\alpha(1-3)$ -linked fucose in part of these glycopeptides, whereas no $\alpha(1-6)$ -linked fucose could be detected in these structures. The degree of β -galactosylation of the complex-type glycans was investigated by chromatog. on Ricinus communis agglutinin-agarose. The results indicated that only part of the biantennary glycans were completely β -galactosylated. Similarly, at least 3 β -galactose residues were present in only a part of the tri-, and tetra- and/or tri'-antennary glycans.

L22 ANSWER 21 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1988:183218 HCPLUS
 DOCUMENT NUMBER: 108:183218
 TITLE: Purification and characterization of calcium-binding conchiolin shell peptides from the mollusc, *Haliotis rufescens*, as a function of development
 Cariolou, Marios A.; Morse, Daniel E.
 AUTHOR(S):
 CORPORATE SOURCE: Mar. Sci. Inst., Univ. California, Santa Barbara, CA, 93106, USA
 SOURCE: Journal of Comparative Physiology, B: Biochemical, Systemic, and Environmental Physiology (1988), 157(6), 717-29
 CODEN: JPBPD; ISSN: 0174-1578

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Conchiolin peptides of the molluscan shell are believed to determine structural organization and facilitate calcification during shell formation. Changes in patterns of conchiolin synthesis during development, and the possible contribution of these peptides to shell formation, have been investigated by purification and characterization of the soluble peptides extracted from the shell

of the gastropod mollusk, *H. rufescens* (red abalone), at various stages of development. Shell peptides were purified from young post-larvae, juveniles, and adults by gel-filtration column chromatog. in aggregation-reducing bicarbonate buffers. Ca-binding domains were detected spectrophotometrically after reaction with a cationic carbocyanine dye. Juvenile and adult shell peptides were heterogeneous, and rich in aspartic acid and glycine residues; in contrast, post-larval shells contained major glycine-rich component. The juvenile shell peptide population shares components from each of the other 2 populations, suggesting that the synthesis of the different shell peptides results from the differential expression of a multi-gene family, in a developmentally controlled progression. Enzymic analyses suggest that Ca binds to the aspartic acid residues of the peptide core, rather than to satellite groups such as phosphate, sulfate, or carbohydrate. The possibility is discussed that the aspartic acid residues found in shell peptides may play an important role in the calcification of the abalone shell matrix. The methods demonstrated here also should prove useful for the purification, characterization, and comparative anal. of Ca-binding proteins of connective tissues, extracellular matrixes and support structures in many other systems.

L22 ANSWER 22 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1987:594390 HCPLUS
 DOCUMENT NUMBER: 107:194390
 TITLE: Purification and properties of prostaglandin E1/prostacyclin receptor of human blood platelets
 AUTHOR(S): Dutta-Roy, Asim K.; Sinha, Asru K.
 CORPORATE SOURCE: Dep. Med., Wright State Univ., Dayton, OH, 45428, USA

SOURCE: Journal of Biological Chemistry (1987), 262(26),
12685-91
CODEN: JBCHA3; ISSN: 0021-9258

DOCUMENT TYPE: Journal
LANGUAGE: English

AB Prostaglandin E1/prostacyclin receptors of human platelets were solubilized in buffer, containing 0.05% Triton X-100 and protease inhibitors. The soluble membrane protein was chromatographed on a DEAE-cellulose column and assayed by a microfiber filter by an equilibrium-binding technique. The active fractions eluted at 0.7M KCl were pooled, and the receptors were purified to homogeneity by Sephadex G-200 gel filtration with an overall recovery of 30%. The isolated receptor was 2200-fold purified over the starting platelets. As evidenced by SDS-PAGE, the receptor showed a mol. mass of 190,000 daltons and is composed of 2 nonidentical subunits with mol. masses of 85,000 and 95,000 daltons. The interaction of prostaglandin E1 with the purified receptor was rapid, saturable, reversible, and highly specific. Among all prostaglandins tested, only prostacyclin was capable of displacing [³H]prostaglandin E1 bound to the receptor. Scatchard anal. of [³H]prostaglandin E1 binding to the purified receptor suggested the presence of a single class of high-affinity binding sites (dissociation constant (K_d) = 9.8 nM) and a 2nd population of low-affinity binding sites (K_d = 0.7 μ M) in the same protein mol. Incubation of the purified receptor with platelets stripped of the receptor by washing with low concns. of Triton X-100 efficiently restored the ability of prostaglandin E1 and prostacyclin to activate adenylate cyclase in these cells.

L22 ANSWER 23 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 1987:454603 HCPLUS
DOCUMENT NUMBER: 107:54603
TITLE: Characterization of an outer membrane mannanase from Bacteroides ovatus
AUTHOR(S): Gherardini, Frank C.; Salyers, Abigail A.
CORPORATE SOURCE: Dep. Microbiol., Univ. Illinois, Urbana, IL, 61801, USA
SOURCE: Journal of Bacteriology (1987), 169(5), 2031-7
CODEN: JOBAAY; ISSN: 0021-9193

DOCUMENT TYPE: Journal
LANGUAGE: English

AB When inner and outer membranes of *B. ovatus* were separated on sucrose gradients, the mannanase activity was associated mainly with fractions containing outer membranes. Enzyme activity was solubilized by 3-[(3-cholamidopropyl)dimethylammonio]-1-propanesulfonate (CHAPS) or by Triton X-100 at a detergent-to-protein ratio of 1:1. The enzyme was stable for only 4 h at 37° and for 50-60 h at 4°. Anal. of the products of the CHAPS-solubilized mannanase on Bio-Gel A-5M and Bio-Gel P-10 gel filtration columns indicated that the enzyme breaks guar gum into high-mol.-weight fragments. The CHAPS-solubilized mannanase was partially purified by chromatog. on a FPLC Mono Q column. The partially purified mannanase preparation contained 3 major polypeptides (mol. weight, Mr 94,500, 61,000, and 43,000) and several minor ones. High mannanase activity was seen only when *B. ovatus* was grown on guar gum. Cross-absorbed antiserum detected 2 other guar gum-associated outer membrane proteins: a CHAPS-extractable 49,000-dalton polypeptide and a 120,000-dalton polypeptide that was not solubilized by CHAPS. Neither of these polypeptides was detectable in the partially purified mannanase preparation. Thus, there are ≥2 guar gum-associated outer

membrane polypeptides other than the mannanase.

L22 ANSWER 24 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1987:435346 HCAPLUS
 DOCUMENT NUMBER: 107:35346
 TITLE: Endogenous lectin from cultured soybean **cells**
 . Chemical characterization of the lectin of SB-1
 cells
 AUTHOR(S): Malek-Hedayat, Shahnaz; Meiners, Sally A.; Metcalf,
 Thomas N., III; Schindler, Melvin; Wang, John L.; Ho,
 Siu Cheong
 CORPORATE SOURCE: Dep. Biochem., Michigan State Univ., East Lansing, MI,
 48824, USA
 SOURCE: Journal of Biological Chemistry (1987), 262(16),
 7825-30
 CODEN: JBCHA3; ISSN: 0021-9258
 DOCUMENT TYPE: Journal
 LANGUAGE: English
 AB A lectin was identified in **cell** line SB-1, originally derived
 from soybean roots. This lectin, referred to as SB-1 lectin, was
 isolated on the basis of its carbohydrate-binding activity
 (affinity **chromatog.** on Sepharose column derivatized with
 N-caproylgalactosamine) and its immunol. cross-reactivity [immunoblotting
 with rabbit antibodies directed against seed soybean agglutinin (SBA)].
 SDS-PAGE and immunoblotting anal. of SB-1 lectin revealed a major
 polypeptide (mol. weight = .apprx.30,000) which comigrated
 with seed SBA. This form of the lectin was observed in fractions purified
 from culture medium of SB-1 **cells** or supernatant fraction of
 SB-1 **cell** suspension after enzymic removal of **cell**
 wall. Exts. of SB-1 **cells** under some other conditions yielded a
 major band (mol. weight = .apprx.60,000) as revealed by SDS-PAGE and
 immunoblotting with rabbit anti-seed SBA; prolonged incubation of these
 samples in the presence of SDS resulted in the appearance of the
 30-kilodalton (kDa) polypeptide. It appears that the 60-kDa band
 represented a highly stable, even under SDS-PAGE conditions, dimeric form
 of the 30-kDa subunit. The SB-1 lectin derived from the culture medium
 was compared with seed SBA by gel **filtration** and by peptide
 mapping after limited **proteolysis**; no difference between the
 lectins from the 2 sources was found. Exts. of soybean roots fractionated
 on N-caproylgalactosamine-Sepharose affinity columns yielded, upon elution
 with galactose, polypeptides of mol. wts. 30,000 and 60,000. The results
 suggested that soybean roots **contain** a lectin whose polypeptide
 composition corresponds to that of seed SBA and SB-1 lectin.

L22 ANSWER 25 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1987:154632 HCAPLUS
 DOCUMENT NUMBER: 106:154632
 TITLE: Purification of **varicella-zoster-virus**
 immunoreactive glycoproteins for kuse as immunoassay
 reagents for **detection** of virus antibodies
 INVENTOR(S): Ellis, Ronald W.; Neff, Beverly J.; Keller, Paul M.;
 Emini, Emilio A.
 PATENT ASSIGNEE(S): Merck and Co., Inc. , USA
 SOURCE: Eur. Pat. Appl., 55 pp.
 CODEN: EPXXDW
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 211756	A1	19870225	EP 1986-401702	19860730
R: AT, BE, CH, DE, FR, GB, IT, LI, LU, NL, SE				
DK 8603642	A	19870320	DK 1986-3642	19860731
JP 62112000	A2	19870522	JP 1986-178992	19860731
ES 2001864	A6	19880701	ES 1986-778	19860731
JP 02022298	A2	19900125	JP 1989-41936	19890223
PRIORITY APPLN. INFO.:			US 1985-761245	19850801
			US 1985-761246	19850801
			US 1985-761248	19850801

AB The glycoproteins gA, gB, and gC of varicella-zoster virus (VZV) are each purified by immunoaffinity chromatog. and are used as diagnostic reagents for detecting antibodies to the virus in biol. fluids. Monoclonal antibodies to gA, gB, and gC are obtained by the hybridoma method according to a known protocol. Individual glycoproteins are purified from MRC-5 human diploid fibroblasts (infected with VZV to 80% cytopathic effect) by immunoaffinity chromatog. using Sepharose 4B containing coupled monoclonal antibodies to gA, gB, or gC. A mixture of the 3 glycoproteins is obtained by purification of the infected-cell extract on a lectin affinity column. Each purified glycoprotein could be immunopptd. only by its homologous monoclonal antibody. The lectin-purified glycoproteins are in their immunol. active form.

L22 ANSWER 26 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1985:74420 HCPLUS
 DOCUMENT NUMBER: 102:74420
 TITLE: Properties of an interphotoreceptor retinoid-binding protein from bovine retina
 AUTHOR(S): Saari, John C.; Teller, David C.; Crabb, John W.; Bredberg, Lucille
 CORPORATE SOURCE: Sch. Med., Univ. Washington, Seattle, WA, 98195, USA
 SOURCE: Journal of Biological Chemistry (1985), 260(1), 195-201
 CODEN: JBCHA3; ISSN: 0021-9258
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB Washes and exts. of frozen and fresh cattle retina contain a water-soluble high-mol. weight, retinoid-binding protein (RBP) that is distinct from 3 other RBPs previously isolated from this tissue and is designated interphotoreceptor retinoid-binding protein (IRBP).. IRBP was purified to apparent homogeneity from retinal homogenates by gel filtration, lectin, and ion-exchange chromatog. Overestimation of the protein mol. weight was observed in several systems involving migration of the protein through a porous network. The approx. mol. weight obtained by SDS-polyacrylamide gel electrophoresis was 140,000, a value consistent with those reported by other labs. However, a more detailed anal., using the method of Ferguson, revealed the protein to behave anomalously relative to several proteins used as SDS-polyacrylamide gel electrophoresis stds. The apparent radius of native IRBP, estimated from calibrated gel filtration, corresponded to a globular protein with a mol. weight of 240,000-280,000, suggesting that the protein is a dimer. However, when the mol. weight of native IRBP was determined by a method with no shape dependence, sedimentation equilibrium, a value of 131,700 g/mol was obtained. Sedimentation equilibrium in a dissociating solvent (6M guanidine-HCl) yielded

a mol. weight for the smallest component of 120,100 g/mol. The similarity of values for the denatured and native mol. weight by sedimentation equilibrium

demonstrated that the protein is a monomer. In further support of this, no evidence for a dimer was observed in crosslinking expts. with di-Me suberimidate. The sedimentation coefficient (5.73 S) and mol. weight from sedimentation equilibrium were employed to calculate the frictional coefficient and

Stokes' radius of IRBP ($f/f_0 = 1.64$, $R_s = 55 \text{ \AA}$, resp.). The high value of f/f_0 of the protein provided a reasonable explanation for the overestn. of the mol. weight of native IRBP on gel **filtration**. Approx. 2 mol of exogenous all-trans- or 11-cis-retinol were bound/mol of protein (131,000 daltons). Approx. 7% of the binding sites were saturated with endogenous ligand (11-cis retinol, 88%; all-trans retinol, 12%) following **isolation** from partially bleached cattle eyes. After more extensive bleaching of cattle retinas, the amount of bound endogenous all-trans retinol increased .apprx.4-fold. The sequence of the 1st 24 amino acids was determined by manual sequence anal. Comparison of this sequence with the N-terminal sequences of 3 other RBPs revealed no obvious similarities. The **extracellular** localization, the complement of endogenous bound retinols, and the capacity to accept addnl. retinol following illumination of the retina were all consistent with the proposed role of the binding protein in the **extracellular** transport of retinol between the retinal pigment epithelium and retina.

L22 ANSWER 27 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1985:58251 HCPLUS

DOCUMENT NUMBER: 102:58251

TITLE: Enzyme antigens associated with pigeon breeder's disease. I. **Isolation** and characterization of basic hydrolases

AUTHOR(S): McCormick, Daniel J.; Tebo, Thomas H.; Calvanico, Nickolas J.; Fredricks, Walter W.

CORPORATE SOURCE: Dep. Biol., Marquette Univ., Milwaukee, WI, USA

SOURCE: Journal of Protein Chemistry (1984), 3(3), 293-308

CODEN: JPCHD2; ISSN: 0277-8033

DOCUMENT TYPE: Journal

LANGUAGE: English

AB A survey of the hydrolytic enzymes present in pigeon dropping exts. (PDE) has shown that this material **contains** a variety of proteolytic and nonproteolytic activities. These enzymes were separated into their basic and acidic components by **chromatog.** on DEAE-cellulose. Staining of immunoppts. with specific chromogenic substrates demonstrated the presence of antibodies in symptomatic breeders to several of the basic enzymes in PDE. Five distinct hydrolytic activities were **isolated** from the basic group of enzymes. Trypsin, elastase, and 2 forms of collagenase were the specific proteolytic activities **isolated**. A phospholipase was also purified from these preps. Purified elastase consisted of a single polypeptide chain (mol. weight 22,000). Purified trypsin had a mol. weight of 25,000 and a charge similar to the 2 reported for elastase. Like elastase, trypsin from PDE was apparently composed of a single polypeptide chain. The 2 mol. weight forms of collagenase both hydrolyzed bovine collagen. High-mol.-weight (51,000) collagenase was a glycoprotein consisting of 2 **polypeptides** (mol. weight 24,000), and was readily separated from low-mol.-weight (15,000) collagenase by gel **filtration**. The phospholipase (mol. weight 99,000) was apparently a dimer. The relevance of these enzymes to the development of pigeon breeder's disease is discussed.

L22 ANSWER 28 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1984:26024 HCPLUS

DOCUMENT NUMBER: 100:26024

TITLE: Urinary antitumor protein

PATENT ASSIGNEE(S): Green Cross Corp., Japan
 SOURCE: Jpn. Kokai Tokkyo Koho, 7 pp.
 CODEN: JKXXAF
 DOCUMENT TYPE: Patent
 LANGUAGE: Japanese
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 58189119	A2	19831104	JP 1982-73887	19820430
PRIORITY APPLN. INFO.:			JP 1982-73887	19820430

AB An antitumor protein (mol. weight 20,000-40,000 dalton; isoelec. point 4.2; maximum absorbance 280 nm; containing α -amino acids) is isolated from the urine of mammals. The preparation inhibited the growth of tumor cells including HeLa cell, KB cell, RT-4 cell, KHM-1 cell, ELC-3 cell, Hep2 cell and MCF 7 cell. Thus, adult urine was passed through a silica gel column (2% NH₄OH as eluent) to give an active fraction, which was adjusted to pH 6-7 and subjected to ultrafiltration. The resultant product was chromatographed on DEAE-cellulose and then Sephadryl 200 and freeze dried.

L22 ANSWER 29 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1983:121619 HCPLUS
 DOCUMENT NUMBER: 98:121619
 TITLE: Isolation of small nuclear ribonucleoproteins containing U1, U2, U4, U5, and U6 RNAs
 AUTHOR(S): Hinterberger, Monique; Pettersson, Ingvar; Steitz, Joan A.
 CORPORATE SOURCE: Dep. Mol. Biophys. Biophys., Yale Univ., New Haven, CT, 06510, USA
 SOURCE: Journal of Biological Chemistry (1983), 258(4), 2604-13
 CODEN: JBCHA3; ISSN: 0021-9258
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB A fractionation procedure was developed for the most abundant small nuclear ribonucleoproteins, the Sm snRNPs containing U1, U2, U4, U5, or U6 RNAs, from mouse Friend erythroleukemia or human HeLa cells. The protocol subjects a nuclear extract to a series of gel filtration, ion exchange, and hydrophobic/ion exchange chromatog. steps. Three final fractions are obtained, 2 of which are essentially U1 snRNPs and a 3rd which contains predominantly the U2 snRNP with minor amts. of U1, U4, U5, and U6 snRNPs. The U RNAs are recovered in .apprx.12% overall yield and comprise >95% of the RNA in the final fractions. They are retained in stable RNA-protein complexes that are completely immunoprecipitable by anti-Sm or anti-(U1)RNP human autoantibodies. Five major polypeptides, with approx. mol. wts. of 11,000, 12,000, 13,000, 16,000, and 28,000 are common to the most highly purified fractions containing either U1 snRNPs or U2, U4, U5, and U6 snRNPs. Three addnl. polypeptides, 22,000, 33,000 and 68,000, are unique to U1 particles; a polypeptide, mol. weight 32,000, is present only in the U2-6 snRNP fraction. The above proteins include all those previously identified as associated with the Sm snRNPs by immunopptn. of fresh cell exts.; they also overlap in size most of the polypeptides reported to carry determinants reactive with anti-Sm or anti-(U1)RNP antisera. The proteins present in the final snRNP preps.

are distinct from histones and from the major proteins of the nuclear matrix or of the nuclear ribonucleoprotein complexes that package mRNA precursors.

L22 ANSWER 30 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1982:558932 HCAPLUS
 DOCUMENT NUMBER: 97:158932
 TITLE: Vacuum-blotting: a new simple and efficient transfer of proteins from sodium dodecyl sulfate-polyacrylamide gels to **nitrocellulose**
 AUTHOR(S): Peferoen, M.; Huybrechts, R.; De Loof, A.
 CORPORATE SOURCE: Zool. Inst., Louvain, 3000, Belg.
 SOURCE: FEBS Letters (1982), 145(2), 369-72
 CODEN: FEBLAL; ISSN: 0014-5793
 DOCUMENT TYPE: Journal
 LANGUAGE: English
 AB An inexpensive, simple, reproducible, and fast vacuum-blotting **method** for polyacrylamide gels and SDS-polyacrylamide gels was developed which uses a vacuum pump **apparatus**, connected to a slab gel dryer system, to transfer electrophoretically separated proteins from the gel onto **nitrocellulose** paper. For large proteins, 0.45- μ -**pore**-size paper is used, and for small **proteins**, **nitrocellulose** with a smaller **pore** size (0.20 or 0.10 μ m) is preferred. After transfer, the proteins are **detected** by a double-antibody immunol. staining technique. The efficiency of the procedure was demonstrated by blotting separated proteins from an egg homogenate and Sarcophaga bullata and from the hemolymph of Leptinotarsa decemlineata.

L22 ANSWER 31 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1982:542937 HCAPLUS
 DOCUMENT NUMBER: 97:142937
 TITLE: Expression of **cell** surface lectins on activated human lymphoid **cells**
 AUTHOR(S): Apgar, John R.; Cresswell, Peter
 CORPORATE SOURCE: Med. Cent., Duke Univ., Durham, NC, USA
 SOURCE: European Journal of Immunology (1982), 12(7), 570-6
 CODEN: EJIMAF; ISSN: 0014-2980
 DOCUMENT TYPE: Journal
 LANGUAGE: English
 AB A **cell** surface lectin found on activated human lymphoid **cells** was identified and characterized using membrane **glycoprotein micelles** as probes. These **micelles**, which are large, water-soluble aggregates, are composed of glycoproteins isolated from detergent-solubilized membranes of human B lymphoblastoid **cell** lines by Lens culinaris hemagglutinin affinity chromatog. The **micelles** have an average apparent mol. weight of 4 + 106 estimated by gel filtration and range in diameter from 25-100 nm. **Micelles** bind to B and T lymphoblastoid **cell** line **cells** and peripheral blood lymphocytes activated with concanavalin A or in a mixed lymphocyte response. Unactivated peripheral blood lymphocytes and red blood **cells** bind very low levels of the **micelles**. The binding is saturable, reversible, and temperature-dependent, with poor binding below 15°. Glycoproteins such as fetuin and porcine thyroglobulin, which contain complex oligosaccharide side chains, inhibit the binding, whereas glycoproteins containing only high mannose or simple serine-linked carbohydrate side chains do not. In addition, binding can be inhibited by complex asparagine-linked glycopeptides purified from pronase-digested fetuin, but not by the simple serine-linked

glycopeptides. Membrane **glycoprotein micelles** are bound to the surface of the **cells** but are not internalized or degraded. The potential role of this **cell** surface lectin in lymphocyte function is discussed.

L22 ANSWER 32 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1982:158904 HCPLUS
 DOCUMENT NUMBER: 96:158904
 TITLE: Outer membrane proteins of **Brucella abortus**:
isolation and characterization
 AUTHOR(S): Verstreate, D. R.; Creasy, M. T.; Caveney, N. T.;
 Baldwin, C. L.; Blab, M. W.; Winter, A. J.
 CORPORATE SOURCE: New York State Coll. Vet. Med., Cornell Univ., Ithaca,
 NY, 14853, USA
 SOURCE: Infection and Immunity (1982), 35(3), 979-89
 CODEN: INFIBR; ISSN: 0019-9567
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB Outer membrane proteins were derived from 1 rough and 4 smooth strains of **B. abortus** by sequential extraction of phys. disrupted **cells** with N-lauroylsarcosinate and dipolar ionic detergent. Extraction of outer membrane proteins was ineffective, however, without predigestion with lysozyme. Three groups of proteins were present and could be separated in their native state by sequential anion-exchange **chromatog.** and gel **filtration**. Membrane proteins **contained** substantial quantities of tightly adherent lipopolysaccharide which could be reduced but not eliminated by extraction of **cells** with TCA before disruption. Group 2 proteins gave rise to 43,000- and 41,000-mol.-weight bands after complete denaturation in SDS. They were antigenically identical among all the strains, showed close resemblance in amino acid **composition** to each other and a general similarity to OmpF of *Escherichia coli*, and are proposed to be the porins of **B. abortus**. Group 3 proteins occurred as 30,000-mol.-weight bands on SDS-polyacrylamide gel electrophoresis, although addnl. bands were frequently observed in this region. In none of the strains did group 3 proteins manifest heat-modifiable characteristics. Proteins of different strains bore a high degree of similarity to each other in amino acid **composition**, except in methionine, isoleucine, tyrosine, and histidine. Differences occurred consistently in amino acid **composition** between group 2 and 3 proteins, and some of these correspond to differences between OmpF and OmpA. Group 2 and 3 proteins were antigenically distinct from each other, but the principal group 3 antigens were shared among all the strains. Despite the lack of heat modifiability, perhaps influenced by adherent lipopolysaccharide, group 3 proteins are proposed as counterparts to OmpA. Most of the group 1 proteins, minor components, were phys. associated with those of group 3 unless in SDS. Group 1 proteins produced a major band at 94,000 and exhibited heat modifiability. No evidence was found of a low-mol.-weight lipoprotein in the outer membrane of **B. abortus**, but this is not taken to exclude its occurrence.

L22 ANSWER 33 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1982:81866 HCPLUS
 DOCUMENT NUMBER: 96:81866
 TITLE: **Isolation** and characterization of peptides obtained by cyanogen bromide cleavage of the β' -subunit of *E. coli* DNA-dependent RNA polymerase
 AUTHOR(S): Shuvaeva, T. M.; Lipkin, V. M.; Nazimov, I. V.;
 Modyanov, N. N.; Ovchinnikov, Yu. A.
 CORPORATE SOURCE: M. M. Shemyakin Inst. Bioorg. Chem., Moscow, USSR

SOURCE: Bioorganicheskaya Khimiya (1981), 7(12), 1765-77
 CODEN: BIKHD7; ISSN: 0132-3423

DOCUMENT TYPE: Journal
 LANGUAGE: Russian

AB The β' -subunit of DNA-dependent RNA polymerase was subjected to CNBr cleavage. BuOH extraction was highly effective for isolating hydrophobic peptides; acetylcellulose slab electrophoresis was used to sep. high mol. weight fragments. By these methods, in combination with gel-filtration and paper or thin-layer chromatog., 21 protein fragments were isolated. Mol. weight and amino acid composition were determined for all isolated compds. In total, these fragments contain .apprx.900 amino acid residues out of .apprx.1450 residues of the β' -subunit. Amino acid sequences were established for a number of peptides.

L22 ANSWER 34 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1981:98312 HCPLUS

DOCUMENT NUMBER: 94:98312

TITLE: Large scale purification and structural characterization of squalene and sterol carrier protein

AUTHOR(S): Dempsey, Mary E.; McCoy, Kim E.; Baker, H. Nordean; Dimitriadou-Vafiadou, A.; Lorsbach, Thomas; Howard, James Bryant

CORPORATE SOURCE: Dep. Biochem., Univ. Minnesota, Minneapolis, MN, 55455, USA

SOURCE: Journal of Biological Chemistry (1981), 256(4), 1867-73
 CODEN: JBCHA3; ISSN: 0021-9258

DOCUMENT TYPE: Journal

LANGUAGE: English

AB A large-scale purification procedure was developed for isolation from rat liver of the protein originally called squalene and sterol carrier protein (SCP). Homogeneous SCP was obtained by gel filtration of the liver soluble proteins on Sephadex G-75 followed by ion-exchange chromatog. on DEAE-cellulose at pH 9.0. SCP represented $\geq 8\%$ of the soluble proteins in the liver extract. SCP functional activity was determined by a spectroscopic assay measuring activation of membrane-bound $\Delta 7$ -sterol $\Delta 5$ -dehydrogenase. Structural studies indicated that SCP is a single polypeptide chain (mol. weight = 16,000; pI = 7.0). SCP had 1 free SH group, partially buried in the native protein. No N-terminal residue was detected by Edman degradation, dansylation, or by Edman degradation after digestion of SCP by pyroglutaminase. The C-terminal sequence of SCP was determined by carboxypeptidase Y degradation. SCP did not contain tryptophan residues or associated carbohydrate, phospholipid, sterol, or phosphate moieties. However, native SCP contained 2 mol of associated fatty acids; 0.5 mol each of palmitic and stearic acids was tightly or covalently attached, whereas similar levels of these acids were noncovalently bound. Apparently, SCP is the fatty acid-binding protein isolated by several groups. A 2nd protein (mol. weight = 14,000; pI = 9.0) was present in the SCP pool from the gel filtration step. This protein was separated from SCP and purified to homogeneity. The 2nd protein was a single polypeptide chain contg . 1 free SH group partially buried in the native protein. As found for SCP, this protein had a blocked N-terminal group not released by pyroglutaminase, contained tightly or covalently bound palmitic and stearic acids, and did not have associated phospholipid, sterol, or phosphate moieties. The 2nd protein, distinguished from SCP by its amino acid composition and peptide maps, did not exhibit SCP functional

activity with membrane-bound enzymes. The abundance of SCP in liver, its ubiquitous occurrence, and broad functions indicate SCP is capable of playing a major role in regulation of lipid metabolism

L22 ANSWER 35 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1980:632892 HCAPLUS
 DOCUMENT NUMBER: 93:232892
 TITLE: Intestinal membrane calcium-binding protein. Vitamin D-dependent membrane component of the intestinal calcium transport **mechanism**
 AUTHOR(S): Kowarski, Szloma; Schachter, David
 CORPORATE SOURCE: Coll. Physicians Surg., Columbia Univ., New York, NY, 10032, USA
 SOURCE: Journal of Biological Chemistry (1980), 255(22), 10834-40
 CODEN: JBCHA3; ISSN: 0021-9258
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB A particulate fraction of rat intestinal mucosal homogenates, termed the Ca-binding complex, **contains** 3 vitamin D-dependent activities: Ca binding of high affinity, Ca-dependent ATPase, and p-nitrophenylphosphatase. These particulate activities vary concordantly with intestinal Ca transport, suggesting that they represent membrane components of the translocation **mechanism**. The particulate was solubilized with 1-BuOH and the activities were resolved partially by gel filtration and by DEAE-cellulose and spheroidal hydroxylapatite column chromatog. The Ca-binding activity was separated from the enzymes and isolated as a protein of mol. weight .apprx.200,000, as estimated by gel filtration in 0.1% Triton X-100. The membrane protein, named IMCal (intestinal membrane Ca-binding protein), was dissociated with SDS to yield a monomer of mol. weight 20,500 which is clearly distinguishable from the soluble Ca-binding protein (mol. weight 11,500) of rat mucosa. The apparent dissociation consts. for Ca²⁺ of IMCal and of the soluble Ca-binding protein were estimated as 0.37 μM and 2.25 μM, resp. The vitamin D-dependent activities of the Ca-binding complex are present in isolated intestinal microvillus membranes and may mediate the translocation of Ca from the intestinal lumen to the cytosol.

L22 ANSWER 36 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1980:468089 HCAPLUS
 DOCUMENT NUMBER: 93:68089
 TITLE: Comparison of glycopeptides from control and virus-transformed baby hamster kidney fibroblasts
 AUTHOR(S): Blithe, Diana L.; Buck, Clayton A.; Warren, Leonard
 CORPORATE SOURCE: Wistar Inst., Philadelphia, PA, 19104, USA
 SOURCE: Biochemistry (1980), 19(14), 3386-95
 CODEN: BICHAW; ISSN: 0006-2960
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB Glucosamine-labeled glycopeptides from control and virus-transformed baby hamster kidney fibroblasts were characterized by size, lectin affinity, charge, and composition. As already demonstrated, on the basis of elution position on a column of Sephadex G-50, transformed cells contained a greater proportion of large glycopeptides than did control cells. Transformed cells also contained a larger proportion of glycopeptides which did not bind to concanavalin A-Sepharose. By sequential chromatog. on Sephadex G-50, concanavalin A-Sepharose, and DEAE-Sephadex, individual peaks were partially or completely resolved. If sialic acid was removed

from the glycopeptides prior to anal. by ion-exchange chromatog ., 95% of the glycopeptides from control **cells** and 85% of the glycopeptides from transformed **cells** were no longer bound by DEAE-Sephadex. It was concluded that the DEAE-Sephadex elution properties of the glycopeptides are determined almost entirely by the sialic acid content of the mols. A comparison of the profiles of control and transformed **cell** glycopeptides simultaneously eluting from columns of DEAE-Sephadex revealed that the differences between the two **cells** were largely quant. There was one component on the surface of transformed **cells** that was virtually absent in control **cells**. It was degraded by nitrous acid **hydrolysis** and heparinase and appeared to be heparan sulfate-like material. After fractionation, each **isolated glycopeptide population** was analyzed for carbohydrate and, in some cases, amino acid content. The apparently larger glycopeptides, group A, the dominant population in transformed **cells**, were found to contain 3 to 4 mannose residues/glycopeptide when the sugars were normalized to sialic acid content. On the basis of the same criteria, group B glycopeptides contained 4-6 mannose residues/glycopeptide. The carbohydrate and amino acid compns. of the glycopeptides from transformed **cells** were, with a few exceptions, similar to those from control **cells** . Some **isolated** glycopeptides appeared to contain both O-glycosidic and N-glycosidic linkages on the same oligopeptide.

L22 ANSWER 37 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1977:514278 HCPLUS
 DOCUMENT NUMBER: 87:114278
 TITLE: Enzymes of the yeast lytic system produced by Arthrobacter GJM-1 bacterium and their role in the lysis of yeast **cell** walls
 AUTHOR(S): Vrsanska, M.; Biely, P.; Kratky, Z.
 CORPORATE SOURCE: Inst. Chem., Sovak Acad. Sci., Bratislava, Czech.
 SOURCE: Zeitschrift fuer Allgemeine Mikrobiologie (1977), 17(6), 465-80
 CODEN: ZAPOAK; ISSN: 0044-2208

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The yeast lytic system produced by Arthrobacter GJM-1 bacterium during growth on bakers' yeast **cell** walls contains a complete set of enzymes which can hydrolyze all structural components of **cell** walls of *Saccharomyces cerevisiae*. Chromatog. fractionation of the lytic system showed the presence of 2 types of endo- β -1,3-glucanase. Rapid lysis of isolated **cell** walls of yeast was induced only by endo- β -1,3-glucanase exhibiting high affinity to insol. β -1,3-glucans and releasing laminaripentaose as the main product of hydrolysis of β -1,3-glucans. This enzyme was able to lyse intact **cells** of *S. cerevisiae* only in the presence of an addition factor present in the Arthrobacter GJM-1 lytic system, which was identified as an alkaline protease. This enzyme possesses the lowest mol. weight among other identified enzyme components present in the lytic system. Its role in the solubilization of yeast **cell** walls from the outer surface by endo- β -1,3-glucanase could be substituted by preincubation of **cells** with Pronase or by allowing the glucanase to act on **cells** in the presence of thiol reagents. The mechanism of lysis of intact **cells** and isolated **cell** walls by the enzymes of Arthrobacter GJM-1 is discussed in the light of the present conception of yeast **cell** wall structure.

L22 ANSWER 38 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1976:132202 HCAPLUS
 DOCUMENT NUMBER: 84:132202
 TITLE: Fluorescence of tryptophan-containing peptides on paper or silica gel after treatment with formaldehyde, formaldehyde-ozone or formaldehyde-hydrochloric acid
 AUTHOR(S): Larsson, L. I.; Sundler, F.; Hakanson, R.
 CORPORATE SOURCE: Dep. Histol., Univ. Lund, Lund, Swed.
 SOURCE: Journal of Chromatography (1976), 117(2), 355-63
 DOCUMENT TYPE: Journal
 LANGUAGE: English
 AB Sensitive and specific procedures for the chromatog. detection of tryptophan and tryptophan-containing peptides are described. H₂CO gas induces strong and characteristic fluorescence from tryptophan and peptides with amino-terminal tryptophan residues on silica gel. On filter-paper, the detection of small amts. of these compds. requires the addnl. use of an oxidant, such as O₃. Treatment with H₂CO-HCl was used as a method for inducing fluorescence from tryptophan-containing peptides regardless of the position of the tryptophan residue in the peptide mol. This reaction is useful for the chromatog. demonstration of small amts. of such peptides on both paper and silica gel. The spectral properties of the fluorophores of such tryptophan-containing peptides are distinctive and serve to distinguish them from all other known biogenic compds. that are capable of giving fluorescence with H₂CO.

L22 ANSWER 39 OF 46 HCAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1976:15501 HCAPLUS
 DOCUMENT NUMBER: 84:15501
 TITLE: Isolation and characterization of a novel vitamin B₁₂-binding protein associated with hepatocellular carcinoma
 AUTHOR(S): Burger, Robert L.; Waxman, Samuel; Gilbert, Harriet S.; Mehlman, Carol S.; Allen, Robert H.
 CORPORATE SOURCE: Sch. Med., Washington Univ., St. Louis, MO, USA
 SOURCE: Journal of Clinical Investigation (1975), 56(5), 1262-70
 DOCUMENT TYPE: Journal
 LANGUAGE: English
 AB High levels of a novel vitamin B₁₂-binding protein (hepatoma B₁₂ BP) were observed recently in plasma obtained from 3 adolescent patients with hepatocellular carcinoma. This protein was isolated in homogeneous form from the plasma and pleural fluid of 2 of these patients by affinity chromatog. with vitamin B₁₂-Sepharose. Hepatoma B₁₂ BP belonged to the R-type group of B₁₂-binding proteins and was essentially indistinguishable from the recently isolated human milk and saliva R-type proteins in terms of: (1) immunol. properties based on immunodiffusion and immunoprecipitation assays; (2) amino acid composition; (3) mol. weight based on amino acid and carbohydrate content; and (4) absorption spectra. Both hepatoma B₁₂ BP's contained more sialic acid and less fucose than the milk and saliva B₁₂ BP's. All 4 proteins contained similar amts. of galactose, mannose, galactosamine, and glucosamine. Differences in sialic acid content accounted for differences in electrophoretic mobility observed among 4 proteins. Differences in total carbohydrate content accounted for the differences in apparent mol. weight observed with both gel filtration and Na dodecyl sulfate-polyacrylamide gel electrophoresis. Tumor tissue from one of the patients contained 10 times as much R-type

protein as did normal liver tissue from the same patient. This suggested that synthesis by the tumor caused the high levels of R-type protein in the plasma of certain patients with **hepatocellular** carcinoma. Plasma survival studies performed with rabbits indicated that the hepatoma B12 BP had a prolonged.

L22 ANSWER 40 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1975:55862 HCPLUS
 DOCUMENT NUMBER: 82:55862
 TITLE: P2 protein of bovine root myelin. **Isolation**
 and some chemical and immunological properties
 AUTHOR(S): Brostoff, S. W.; Sacks, H.; Dal Canto, M.; Johnson, A.
 B.; Raine, C. S.; Wisniewski, H.
 CORPORATE SOURCE: Dep. Pathol., Albert Einstein Coll. Med., Bronx, NY,
 USA
 SOURCE: Journal of Neurochemistry (1974), 23(5), 1037-43
 CODEN: JONRA9; ISSN: 0022-3042
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB A small basic **protein** (**mol.** weight 12,000) i.e. the P2 protein, was extracted with dilute acid from delipidated bovine spinal nerve root myelin and purified by ion exchange **chromatog.** on **cellulose** phosphate. It appeared homogeneous on polyacrylamide gel electrophoresis. The P2 protein had a distinctly different amino acid **composition** than the larger basic **protein** (**mol.** weight 18,000), i.e. P1 protein, that is also present in peripheral nerve myelin. It **contained** relatively more hydrophobic residues and much less histidine and proline. The P2 protein conjugated with peroxidase was bound by lymph node **cells** and **infiltrates** in rabbits sensitized with whole bovine root myelin. No binding was evident with the bovine central nervous system myelin basic protein. Chemical and immunol., the P2 protein appeared specific to peripheral nervous system myelin. The **isolated** P2 protein produced mild clin. symptoms of exptl. allergic neuritis, but no histol. evidence of disease. It was suggested that the P2 protein is an important antigen for exptl. allergic neuritis, and that its antigenic determinants may be conformation-dependent.

L22 ANSWER 41 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1972:471698 HCPLUS
 DOCUMENT NUMBER: 77:71698
 TITLE: Further characterization of the alkali-stable material
 from the scales of *Pleurochrysis scherffelii*.
 AUTHOR(S): Cellulosic glycoprotein
 Herth, Werner; Franke, Werner W.; Stadler, Joachim;
 Bittiger, Helmut; Keilich, Gunda; Brown, R. Malcolm,
 Jr.
 CORPORATE SOURCE: Inst. Biol. II, Univ. Freiburg/Br., Freiburg/Br., Fed.
 Rep. Ger.
 SOURCE: Planta (1972), 105(1), 79-92
 CODEN: PLANAB; ISSN: 0032-0935
 DOCUMENT TYPE: Journal
 LANGUAGE: English
 AB Crude and alkali-purified fractions of scales **isolated** from the haptophycean alga *P. scherffelii* were studied. The fibrillar, alkali-resistant polysaccharide component was shown to be predominantly **cellulose** (I) by gas **chromatog.** of the hydrolyzate. Glucose (α - and β -) was the predominant sugar monomer with some galactose and pentoses and the disaccharide **cellobiose**. X-ray diffraction diagrams corresponded to those of **cellulose** II, indicating that I was the main crystalline component. Nitration yielded I

nitrate and indicated that 15.1% of the dry weight was I. The degree of polymerization by viscosity was 3150, but the distribution of chain lengths was very heterogeneous. Scale I was more resistant to alkaline and acid hydrolysis than cotton and wood I. A peptide was covalently linked to the polysaccharide structure and remained associated with it during the purification procedures. The protein content of the alkali-purified scale material was 32% dry weight indicating a cellulose:protein (weight:weight) of approx. 2:1. The peptide contained Asn, threonine, serine (II), Gln, glycine, alanine, valine, isoleucine, and leucine with II as the major amino acid. Cellulase, lysozyme, trypsin, and pronase did not degrade the scale fibrils. It was concluded that fibrils consisting of a β -(1 → 4) glucan were produced within the dictyosomal cisternae of Pleurochrysis with crystals characteristic of I. The relation of this cellulolic glycoprotein to other cell wall glycoproteins was discussed and a hypothetical pathway for scale production was proposed.

L22 ANSWER 42 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1971:9635 HCPLUS

DOCUMENT NUMBER: 74:9635

TITLE: Autolysis of isolated cell

walls of *Bacillus licheniformis* NCTC 6346 and *Bacillus subtilis* Marburg strain 168. Separation of the products and characterization of the mucopeptide fragments

AUTHOR(S): Hughes, Reginald C.

CORPORATE SOURCE: Nat. Inst. Med. Res., London, UK

SOURCE: Biochemical Journal (1970), 119(5), 849-60

CODEN: BIJOAK; ISSN: 0264-6021

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Cell walls were isolated from *B. licheniformis* NCTC 6346 and *B. subtilis* Marburg strain 168 trp grown on casein hydrolysate into the exponential phase. Autolysis was carried out and the soluble products, separated by chromatog. on DEAE-cellulose, from the 2 wall preps. are broadly similar in composition and are in agreement with autolysis proceeding with hydrolysis of amide bonds between L-alanine and N-acetylmuramic acid residues in the mucopeptide components. Peptides originating from the mucopeptide components were isolated and shown to be a monomer peptide, L-alanyl-D-glutamyl-meso-diaminopimelic acid and a dimer peptide containing 2 monomer peptides linked through a residue of D-alanine. Approx. 1 amide group is present for each equivalent tripeptide unit and is probably substituted on diaminopimelic acid residues. Oligosaccharides originating from the mucopeptide components were isolated and after hydrolysis contained almost equimolar amts. of glucosamine and muramic acid and only small amts. of amino acids. The number-average chain length, estimated by the release of

non-reducing end groups of N-acetylglucosamine with exo- β -N-acetylglucosaminidase, is .apprx.10 hexosamine residues for oligosaccharides isolated from either organism. The oligosaccharides are polydisperse. N-Acetylglucosamine residues are the only reducing terminals detectable in the oligosaccharides isolated from *B. subtilis* or *B. licheniformis* cell-wall autolysates. The number-average chain lengths of the oligosaccharides were determined by estimation of the content of these residues and are higher than those found by enzymic assay.

L22 ANSWER 43 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1969:9424 HCPLUS
 DOCUMENT NUMBER: 70:9424
 TITLE: Isolation of chromatographically pure toxin of Clostridium botulinum type B
 AUTHOR(S): DasGupta, Bibhuti R.; Boroff, Daniel A.; Cheong, Kathleen
 CORPORATE SOURCE: Albert Einstein Med. Center, Philadelphia, PA, USA
 SOURCE: Biochemical and Biophysical Research Communications (1968), 32(6), 1057-63
 CODEN: BBRCA9; ISSN: 0006-291X
 DOCUMENT TYPE: Journal
 LANGUAGE: English
 AB C. botulinum type B toxin was isolated, purified, and analyzed on a DEAE-cellulose chromatographic column. At least 4 components were found, 1 toxic and the other 3 nontoxic or slightly toxic. A toxin from C. botulinum type B, strain Lamanna was also isolated and chromatographed; 3 peaks were obtained. The first peak (G-100) was combined with solid $(\text{NH}_4)_2\text{SO}_4$ and the precipitate contained the toxin. Repeated rechromatog. of the toxin gave a $\text{B}\alpha$ fraction which was examined for homogeneity by gel filtration. Only 1 sym. peak was eluted. $\text{B}\alpha$ is a simple protein, mol. weight 165,000. The method described resulted in a 500 fold purification of the toxin with a 13-15% yield of total toxic activity in the bacterial culture. The sp. toxicities of types B and A, expressed as MLD/1.0 absorbance at 278 μm , were 5.9 + 107 and 9 + 107, resp. The mol. weight of $\text{B}\alpha$ is higher than $\text{A}\alpha$ (150,000), and $\text{B}\alpha$ is a more anionic protein on a DEAE-Sephadex column than $\text{A}\alpha$.

L22 ANSWER 44 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1968:400826 HCPLUS
 DOCUMENT NUMBER: 69:826
 TITLE: Designation of high-molecular substances accumulating in the medium during the growth of Scenedesmus obliquus
 AUTHOR(S): Burczyk, Jan
 CORPORATE SOURCE: Inst. Zootech., Grodziec Slaski, Pol.
 SOURCE: Folia Biologica (Krakow, Poland) (1968), 16(1), 55-66
 CODEN: FOBGA8; ISSN: 0015-5497
 DOCUMENT TYPE: Journal
 LANGUAGE: English
 AB Substances of mol. weight >100,000 were isolated, using gel filtration, from the cooled growth medium of S. obliquus treated with a large excess of MeOH. The substances were polarographically active, and their concentration in the medium increased with increasing nos. of algae in the culture. Determination of N, gel electrophoresis, uv spectroscopy, and acid hydrolysis followed by paper chromatog. indicated that the substances contained mucoproteins with at least 14 amino acids. The sterilized substances added to fresh cultures inhibited growth slightly, especially in the logarithmic growth phase. Similar inhibition was noted under N-starved conditions. The source of the substances appeared to be broken cell walls from the algae.

L22 ANSWER 45 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN
 ACCESSION NUMBER: 1967:505286 HCPLUS
 DOCUMENT NUMBER: 67:105286
 TITLE: Fractionation of isoacceptor transfer RNAs on protein-cellulose columns

AUTHOR(S): Artamonova, V. A.; Frolova, L. Yu.; Kiselev, L. L.
 CORPORATE SOURCE: Inst. Epidemiol. Mikrobiol. im. Gamalei, Moscow, USSR
 SOURCE: Dent. Cosmos (1967), 1(4), 530-8
 DOCUMENT TYPE: Journal
 LANGUAGE: Russian

AB **Aminocellulose** prepared according to Gurvich, et al., (CA 53: 9438i) was stirred for 20 min. at 0° in a solution containing 2% NaNO₂ and 5% HCl. The diazotized **cellulose** was washed with H₂O and mixed with a solution of histone or protamine in 0.1M borate buffer (pH 8.6) for 18-20 hrs. in the cold. The sediment was washed with borate buffer and H₂O, and dried. If the weight ratio of the added protein to diazotized **cellulose** was 1:2-4, the resulting complex contained about 2 or 3 weight % of histone or protamine, resp. After preliminary washing with 2M NaCl, H₂O, and 0.025M citrate buffer (pH 5.5) the columns (0.8 + 6 or 1.2 + 8 cm.) were packed and washed with the buffer. The ¹⁴C-labeled aminoacyl RNAs isolated from yeast or rat liver (Kiseleva and Frolov, CA 62: 8041d) were fractionated in the cold in a linear (0.2-1.5M) or step-wise gradient of NaCl in the buffer. The resolution of the fractions was improved if the peaks were rechromatographed or if oligonucleotides bound to aminoacyl residue were fractionated. The aminoacyl oligonucleotides were obtained by hydrolysis with guanyl RNase (I) (Grachev, et al., CA 64: 8537g). Three serine-specific and 3 valine-specific fractions were found in the yeast transfer RNA (tRNA) and 1 valine-specific fraction and 4 serine-specific fractions in rat liver tRNA, if both were hydrolyzed with rat liver I.

L22 ANSWER 46 OF 46 HCPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER: 1966:415906 HCPLUS
 DOCUMENT NUMBER: 65:15906
 ORIGINAL REFERENCE NO.: 65:2977c-d
 TITLE: Electrophoresis
 AUTHOR(S): Strickland, R. D.
 CORPORATE SOURCE: Veterans Admin. Hosp., Albuquerque, NM
 SOURCE: Anal. Chem. (1966), 38(5), 99R-130R
 CODEN: ANCHAM; ISSN: 0003-2700
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB A review of literature primarily from the second half of 1963 through the early part of 1965. Topics covered are fundamental developments, apparatus, stabilizing media, buffers, procedures (general methodology, manipulative techniques, detection and measurement), biol. applications (interactions, biol. fluids, human serum, mammalian serum, lipoproteins, glycoproteins, hemoglobin, specialized blood proteins, cell and particle electrophoresis, tissues, lower vertebrates, invertebrates, taxonomy, plants, microorganisms, enzymes, hormones, biochemicals, and applications to pharmacology and toxicology), and general chemical applications. 2484 references.

=> d que stat 125

L14	25934 SEA FILE=HCAPLUS ABB=ON	(?PROTEIN? OR ?PEPTIDE?) (W) (?MOLECUL?
	OR ?POPUL? OR ?CELL?)	
L15	5686 SEA FILE=HCAPLUS ABB=ON	L14 AND (?ISOLAT? OR ?DETECT?)
L16	2254 SEA FILE=HCAPLUS ABB=ON	L15 AND (?APPARATUS? OR ?MECHANISM?
	OR ?EQUIP? OR ?CONTAIN?)	
L17	979 SEA FILE=HCAPLUS ABB=ON	L16 AND (?PRODUCT? OR ?COMPOS? OR
	?METHOD? OR KIT?)	
L18	303 SEA FILE=HCAPLUS ABB=ON	L17 AND (?LYSIS? OR ?FILT?)
L19	167 SEA FILE=HCAPLUS ABB=ON	L18 AND ?CELL?
L20	4 SEA FILE=HCAPLUS ABB=ON	L19 AND ?PORE?
L21	42 SEA FILE=HCAPLUS ABB=ON	L19 AND ?CHROMATOG?
L22	46 SEA FILE=HCAPLUS ABB=ON	L20 OR L21
L23	221 SEA L22	
L24	200 DUP REMOV L23 (21 DUPLICATES REMOVED)	
L25	24 SEA L24 AND (?RAPID? OR ?QUICK?)	

L25 ANSWER 1 OF 24 MEDLINE on STN
 ACCESSION NUMBER: 2002500138 MEDLINE
 DOCUMENT NUMBER: 22248727 PubMed ID: 12361711
 TITLE: Identification of S-glutathionylated **cellular**
 proteins during oxidative stress and constitutive
 metabolism by affinity purification and proteomic
 analysis.
 AUTHOR: Lind Christina; Gerdes Robert; Hammell Ylva;
 Schuppe-Koistinen Ina; von Lowenhielm Helena Brockenhuis;
 Holmgren Arne; Cotgreave Ian A
 CORPORATE SOURCE: Division of Biochemical Toxicology, Institute of
 Environmental Medicine, Karolinska Institute, Stockholm,
 Sweden.
 SOURCE: ARCHIVES OF BIOCHEMISTRY AND BIOPHYSICS, (2002 Oct 15) 406
 (2) 229-40.
 Journal code: 0372430. ISSN: 0003-9861.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 200211
 ENTRY DATE: Entered STN: 20021004
 Last Updated on STN: 20021213
 Entered Medline: 20021119

AB Redox modification of proteins is proposed to play a central role in regulating **cellular** function. However, high-throughput techniques for the **analysis** of the redox status of individual proteins in complex mixtures are lacking. The aim was thus to develop a suitable technique to **rapidly** identify proteins undergoing oxidation of critical thiols by S-glutathionylation. The **method** is based on the specific reduction of mixed disulfides by glutaredoxin, their reaction with N-ethylmaleimide-biotin, affinity purification of tagged proteins, and identification by proteomic **analysis**. The **method** unequivocally identified 43 mostly novel **cellular** protein substrates for S-glutathionylation. These include protein chaperones, cytoskeletal **proteins**, **cell** cycle regulators, and enzymes of intermediate metabolism. Comparisons of the patterns of S-glutathionylated proteins extracted from **cells** undergoing diamide-induced oxidative stress and during constitutive metabolism reveal both common protein substrates and substrates failing to undergo enhanced S-glutathionylation during oxidative stress. The ability to chemically tag, select, and identify S-glutathionylated proteins, particularly during constitutive metabolism, will greatly enhance efforts

to establish posttranslational redox modification of **cellular** proteins as an important biochemical control **mechanism** in coordinating **cellular** function.

L25 ANSWER 2 OF 24 MEDLINE on STN
 ACCESSION NUMBER: 1998218738 MEDLINE
 DOCUMENT NUMBER: 98218738 PubMed ID: 9538195
 TITLE: Purification and some characteristics of phosphatase of a psychrophile.
 AUTHOR: Tsuruta H; Tsuneta S T; Ishida Y; Watanabe K; Uno T; Aizono Y
 CORPORATE SOURCE: Department of Biofunctional Chemistry, Faculty of Agriculture, Kobe University, Hyogo.
 SOURCE: JOURNAL OF BIOCHEMISTRY, (1998 Feb) 123 (2) 219-25.
 Journal code: 0376600. ISSN: 0021-924X.
 PUB. COUNTRY: Japan
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199808
 ENTRY DATE: Entered STN: 19980828
 Last Updated on STN: 19980828
 Entered Medline: 19980818

AB The phosphatase of a psychrophile was purified by ammonium sulfate fractionation, and a sequence of **chromatographies** on DEAE-**Cellulofine**, butyl-**Cellulofine**, Sephadryl S-100, and Mono-Q columns. The purified enzyme preparation was found to be electrophoretically homogeneous on native- and SDS-PAGE, and its molecular mass was determined to be 38.4 kDa by MALDI-TOF mass spectrometry. Maximal activity was observed at 30 degrees C and pH 6.0. Furthermore, the activity of this enzyme at 0 and 5 degrees C was 27 and 28%, respectively, of that at 30 degrees C. The enzyme was stable in the pH range of 6.0 to 8.0 and up to 20 degrees C. The enzyme was affected by metal ions; the activity was enhanced by Mg²⁺ and Ca²⁺ ions, but depressed by Zn²⁺ ions. **Analysis** of the amino acid **composition** indicated that this phosphatase **contains** no S-S bond, and only a few prolyl residues necessary to retain the rigid structure of a **protein molecule**. The phosphatase shows typical features of a cold enzyme; high catalytic activity at low temperature and **rapid** inactivation at an intermediate temperature.

L25 ANSWER 3 OF 24 MEDLINE on STN
 ACCESSION NUMBER: 93042733 MEDLINE
 DOCUMENT NUMBER: 93042733 PubMed ID: 1420926
 TITLE: Adenine nucleotide translocase greatly increases the partition of trinitrophenyl-ATP into reduced Triton X-100 **micelles**.
 AUTHOR: Tummino P J; Gafni A
 CORPORATE SOURCE: Institute of Gerontology, University of Michigan, Ann Arbor 48109.
 CONTRACT NUMBER: AG 09761 (NIA)
 T32 AG00114 (NIA)
 SOURCE: BIOPHYSICAL JOURNAL, (1992 Oct) 63 (4) 1071-80.
 Journal code: 0370626. ISSN: 0006-3495.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199212
 ENTRY DATE: Entered STN: 19930122

Last Updated on STN: 19970203
 Entered Medline: 19921211

AB The presence of adenine nucleotide translocase (ANT) was found to greatly enhance the partitioning of the ATP analog 2',3'-O-(2,4,6-trinitrophenyl)-adenosine 5'-triphosphate (TNP-ATP) into reduced Triton X-100 **micelles**. The protein's effect was studied through the quenching of fluorescence of purified ANT, irreversibly inhibited by carboxyatractyloside (CAT), solubilized in reduced Triton X-100 **micelles**. The dependence of quenching of the protein's time-resolved tryptophan fluorescence on TNP-ATP concentration was measured and found to follow a Stern-Volmer **mechanism**. However, the calculated quenching constant was too large to be accounted for by the aqueous TNP-ATP concentration. Experiments were therefore conducted to determine the partitioning of the quencher between the three phases present: aqueous, protein-free **micelle**, and **protein-micelle**; a system also described by the equation of Omann, G. M., and M. Glaser (1985. Biophys. J. 47:623-627.). By measuring the dependence of the apparent quenching rate constant on the protein concentration and **protein/micelle** ratios, this equation was used to calculate both the quencher partition coefficient into protein-free **micelles** (P_m) and into **protein-micelles** (P_{pm}), as well as the bimolecular quenching rate constant (k_{pm}) in **protein-micelles**. From the quenching experiments, $k_{pm} = 5.0 \times 10(8) M^{-1}s^{-1}$, $P_m = 290$ and pyrene quenching experiment to be 325, and by a **rapid filtration** experiment to be 450. Clearly, the presence of the integral membrane protein ANT-CAT in reduced Triton X-100 **micelles** greatly increases the partition of TNP-ATP into the **micelle**. ANT alters the properties and thus, the structure of the detergent **micelle**, which has direct implications for the use of detergent **micelles** as a model system for membrane proteins and may indicate that analogous effects occur in the mitochondrial membrane.

L25 ANSWER 4 OF 24 MEDLINE on STN
 ACCESSION NUMBER: 92015504 MEDLINE
 DOCUMENT NUMBER: 92015504 PubMed ID: 1656087
 TITLE: cis- and trans-cleavage activities of poliovirus 2A protease expressed in Escherichia coli.
 AUTHOR: Alvey J C; Wyckoff E E; Yu S F; Lloyd R; Ehrenfeld E
 CORPORATE SOURCE: Department of Cellular Biology, University of Utah, School of Medicine, Salt Lake City 84132.
 CONTRACT NUMBER: AI 12387 (NIAID)
 AI 27914 (NIAID)
 SOURCE: JOURNAL OF VIROLOGY, (1991 Nov) 65 (11) 6077-83.
 Journal code: 0113724. ISSN: 0022-538X.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199111
 ENTRY DATE: Entered STN: 19920124
 Last Updated on STN: 20000303
 Entered Medline: 19911114

AB The poliovirus protease, 2Apro, was produced in Escherichia coli from plasmids that encode a fusion protein consisting of the N-terminal portion of the bacterial TrpE protein linked to poliovirus 2Apro. This fusion protein underwent efficient autocatalytic cleavage at the N terminus of 2Apro, generating the mature protease. Extracts of bacteria expressing 2Apro induced the specific cleavage of the p220 subunit of the eukaryotic translation initiation factor 4F, similar to the 2Apro-mediated reaction

that occurs in poliovirus-infected HeLa cells. A portion of the poliovirus polyprotein containing the 2Apro cleavage site at the P1/P2 junction was produced by translation of cDNA transcripts in rabbit reticulocyte lysates and then tested as a substrate for 2Apro-mediated cleavage. The protein was partially cleaved by 2Apro in trans. Finally, a 16-amino-acid synthetic peptide, representing the P1/P2 junction sequence, was analyzed as a substrate for 2Apro. The peptide was labeled with fluorescein at a lysine residue to facilitate its detection. Recombinant 2Apro cleaved the synthetic peptide into two half-peptide molecules which were resolved by high-pressure liquid chromatography. Direct sequence analysis of the isolated peptide products demonstrated that cleavage occurred at the expected tyrosine-glycine pair. A rapid cleavage assay for 2Apro activity on the synthetic peptide was developed, using separation of the fluorescein-labeled 8-amino-acid product from the 16-residue substrate by electrophoresis on sodium dodecyl sulfate-polyacrylamide gels.

L25 ANSWER 5 OF 24 MEDLINE on STN
 ACCESSION NUMBER: 87308303 MEDLINE
 DOCUMENT NUMBER: 87308303 PubMed ID: 2887571
 TITLE: Purification and properties of prostaglandin E1/prostacyclin receptor of human blood platelets.
 AUTHOR: Dutta-Roy A K; Sinha A K
 SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1987 Sep 15) 262 (26)
 12685-91.
 Journal code: 2985121R. ISSN: 0021-9258.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 198710
 ENTRY DATE: Entered STN: 19900305
 Last Updated on STN: 19980206
 Entered Medline: 19871021

AB Activation of platelet adenylate cyclase by prostaglandin E1 or prostacyclin is initiated through the interaction of the agonists with the same receptors on membrane. Prostaglandin E1/prostacyclin receptors of human platelets were solubilized in buffer, containing 0.05% Triton X-100 and protease inhibitors. The soluble membrane protein was chromatographed on a DEAE-cellulose column and assayed by a microfiber filter by equilibrium binding technique. The active fractions eluted at 0.7 M KCl were pooled, and the receptors were purified to homogeneity by Sephadex G-200 gel filtration with an overall recovery of 30%. The isolated receptor was 2,200-fold purified over the starting platelets. As evidenced by sodium dodecyl sulfate-polyacrylamide gel electrophoresis, the receptor showed a molecular mass of 190,000 daltons and is composed of two nonidentical subunits with molecular masses of 85,000 and 95,000 daltons. The interaction of prostaglandin E1 with the purified receptor was rapid, saturable, reversible, and highly specific. Among all prostaglandins tested, only prostacyclin was capable of displacing [³H]prostaglandin E1 bound to the receptor. Scatchard analysis of [³H]prostaglandin E1 binding to the purified receptor suggested the presence of a single class of high affinity binding sites ($K_d = 9.8 \text{ nM}$) and a second population of low affinity binding sites ($K_d = 0.7 \text{ microM}$) in the same protein molecule. Incubation of the purified receptor with platelets stripped of the receptor by washing with low concentrations of Triton X-100 efficiently restored the ability of prostaglandin E1 and prostacyclin to activate adenylate cyclase in these

cells.

L25 ANSWER 6 OF 24 MEDLINE on STN
 ACCESSION NUMBER: 86205980 MEDLINE
 DOCUMENT NUMBER: 86205980 PubMed ID: 3458213
 TITLE: Identification of the macrophage mannose receptor as a 175-kDa membrane protein.
 AUTHOR: Wileman T E; Lennartz M R; Stahl P D
 CONTRACT NUMBER: AI20015 (NIAID)
 SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1986 Apr) 83 (8) 2501-5.
 Journal code: 7505876. ISSN: 0027-8424.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 198605
 ENTRY DATE: Entered STN: 19900321
 Last Updated on STN: 19970203
 Entered Medline: 19860523

AB Mannose-lactoperoxidase, a neoglycoprotein prepared by reaction of lactoperoxidase with cyanomethyl 1-thiomannoside, bound to alveolar macrophages at 4 degrees C ($K_d = 5.8 \times 10^{-8}$ M) and was **rapidly** internalized at 37 degrees C ($K_u = 2 \times 10^{-8}$ M).
 Mannose-lactoperoxidase binding and uptake were blocked by yeast mannan, and mannose-lactoperoxidase inhibited uptake of ^{125}I -labeled mannose-BSA (bovine serum albumin). Radioiodination of **cells** with surface-bound mannose-lactoperoxidase was carried out in the presence of glucose and glucose oxidase. A major polypeptide (175 kDa) was radioiodinated by this procedure. Iodination of the 175-kDa polypeptide appeared to be receptor-mediated, since it was blocked by the presence of yeast mannan. Specific iodination was absent from receptor-negative **cells**. To demonstrate that the 175-kDa species is a ligand-binding **protein**, **cells** were iodinated by the standard lactoperoxidase **method**. Washed **cells** were then allowed to bind mannose-BSA. Receptor-ligand complexes, prepared by detergent extraction, were passed over anti-BSA IgG affinity columns. Mannose, but not mannose 6-phosphate or galactose, eluted a radioactive protein from the column that migrated with an apparent molecular mass of 175 kDa on NaDODSO₄/PAGE. Detergent extracts of crude membranes prepared from macrophage-enriched whole rabbit lung were adsorbed to mannose-Sepharose; the fraction obtained by elution with mannose contained two protein components of 175 and 55 kDa. Subsequent chromatography on N-acetylglucosamine-agarose yielded a single protein of 175 kDa. The 175-kDa polypeptide was shown to bind ^{125}I -labeled mannose-BSA in a precipitation assay. This binding could be blocked with mannan or mannose-BSA. The results indicate that the **cell**-surface mannose receptor is a 175-kDa protein.

L25 ANSWER 7 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
 ACCESSION NUMBER: 2003:155162 BIOSIS
 DOCUMENT NUMBER: PREV200300155162
 TITLE: **Analysis** of Rabbit Tear Proteins by High Pressure Liquid **Chromatography**-Electrospray Ionization-Mass Spectrometry (LC-ESI-MS).
 AUTHOR(S): Zhou, L. [Reprint Author]; Beuerman, R. W.; Barathi, A. [Reprint Author]; Tan, D.
 CORPORATE SOURCE: Singapore Eye Research Institute, Singapore, Singapore
 SOURCE: ARVO Annual Meeting Abstract Search and Program Planner,

(2002) Vol. 2002, pp. Abstract No. 3140. cd-rom.
 Meeting Info.: Annual Meeting of the Association For
 Research in Vision and Ophthalmology. Fort Lauderdale,
 Florida, USA. May 05-10, 2002.

DOCUMENT TYPE: Conference; (Meeting)
 LANGUAGE: English
 ENTRY DATE: Entered STN: 26 Mar 2003
 Last Updated on STN: 26 Mar 2003

AB Purpose: Tears, a complex protein mixture, are critical to good vision. In this study, we have evaluated the applicability of LC-ESI-MS to display the protein profile from rabbit tears. The response of the tear protein profile to corneal wound healing in a rabbit model was investigated. **Methods:** Tears were collected from New Zealand White rabbits prior to and daily for four days following a unilateral 5mm dia. epithelial wound using 10 mul calibrated glass microcapillary tubes. Tear proteins were eluted by a reverse-phase HPLC column and the tear protein profile was monitored by ESI positive TIC (Total Ion Current) **chromatography.** Results: Tear proteins could be separated into 18 peaks, each of which **contained** a number of protein components. The molecular size of each protein component was determined by on line electrospray ionization (ESI) mass spectrometer. Major tear protein components, lactoferrin, lysozyme (minimally **detectable** in rabbit tears), albumin, lipocalin, lipophilin and beta2-microglobulin were **rapidly** identified by this **method.** Based on the mass data, beta2-microglobulin revealed glycosylation with N-acetyl-hexsamine. ESI positive TIC **chromatograms** and mass spectra indicated differences in tear protein profile before and after corneal wounding. After the cornea was wounded for 24 hours, the level of a protein with molecular weight of 14717 Da was found to be 7-fold higher than that in control tears. It dropped back to normal levels 96-hours after wounding. Conclusion: The LC-ESI-MS has been demonstrated to be a reproducible, fast and simple **method** for identification and **analysis** of many protein components of tears. Importantly, this technique also allows quantification of each component resolved in the **chromatogram.** This **method** is very suitable for mapping peptides and small proteins (< 80 kDa) in tears and should be useful to examine tear protein levels in experimental condition.

L25 ANSWER 8 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
 ACCESSION NUMBER: 2002:412617 BIOSIS
 DOCUMENT NUMBER: PREV200200412617
 TITLE: Capillary **chromatography**-coupled mass spectrometry with column switching for **rapid** identification of proteins from 2-dimensional electrophoresis gels.
 AUTHOR(S): Delinsky, David C.; Greis, Kenneth D. [Reprint author]
 CORPORATE SOURCE: Procter and Gamble Pharmaceuticals, 8700 Mason-Montgomery Road, Mason, OH, 45040, USA
 greis.kd@pg.com
 SOURCE: Journal of Proteome Research, (May-June, 2002) Vol. 1, No. 3, pp. 279-284. print.
 ISSN: 1535-3893.
 DOCUMENT TYPE: Article
 LANGUAGE: English
 ENTRY DATE: Entered STN: 31 Jul 2002
 Last Updated on STN: 31 Jul 2002
 AB An improved capillary liquid **chromatography** procedure, incorporating column switching in combination with mass spectrometry, is reported. The dual column system allows for **rapid**

inject-to-inject cycle times to improve the speed of protein identification for proteomics applications. Full gradient elution of peptides from either of the two C18 columns can be achieved in less than 17 min while maintaining sufficient resolution for the peptides to be detected and fragmented by the mass spectrometer for protein identification. Importantly, the use of two columns for subsequent injections is reproducible and without carry-over. The limit of detection for the system is between 25 and 50 fmol per injection. This fully automated system is capable of analyzing and identifying proteins from an entire 96-well plate in about 27 h.

L25 ANSWER 9 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
 ACCESSION NUMBER: 2002:149828 BIOSIS
 DOCUMENT NUMBER: PREV200200149828
 TITLE: The cytoplasmic tail of L-selectin interacts with members of the Ezrin-Radixin-Moesin (ERM) family of proteins. Cell activation-dependent binding of Moesin but not Ezrin.
 AUTHOR(S): Ivetic, Aleksandar [Reprint author]; Deka, Jurgen; Ridley, Anne; Ager, Ann
 CORPORATE SOURCE: Ludwig Institute for Cancer Research, 91 Riding House St., London, W1W 7BS, UK
 SOURCE: Journal of Biological Chemistry, (January 18, 2002) Vol. 277, No. 3, pp. 2321-2329. print.
 CODEN: JBCHA3. ISSN: 0021-9258.
 DOCUMENT TYPE: Article
 LANGUAGE: English
 ENTRY DATE: Entered STN: 14 Feb 2002
 Last Updated on STN: 26 Feb 2002
 AB L-selectin regulates the recruitment of naive lymphocytes from the bloodstream to secondary lymphoid organs, mediating their initial capture and subsequent rolling along high endothelial cell surface-expressed ligands in peripheral lymph nodes. In vivo, distribution of L-selectin and cell surface levels determine the tethering efficiency and rolling velocity of leukocytes, respectively. Treatment of naive lymphocytes with phorbol myristate acetate (PMA) induces rapid ectodomain proteolytic down-regulation (shedding) of surface L-selectin via a protein kinase C (PKC)-dependent pathway. In an attempt to isolate proteins that are involved in regulating L-selectin expression, an affinity column was constructed using the 17-amino acid cytoplasmic tail of L-selectin. Affinity purification of extracts from lymphocytes, pre-treated with or without PMA, allowed identification of proteins that interact with the affinity column under one condition but not the other. By using this approach, members of the Ezrin-Radixin-Moesin family of proteins were found to interact specifically with the cytoplasmic tail of L-selectin. Moesin from PMA-stimulated lymphocytes, but not from unstimulated lymphocytes, bound to L-selectin tail. In contrast, ezrin from unstimulated or PMA-stimulated lymphocytes associated with L-selectin tail with equal affinity. Furthermore, the PKC inhibitor Ro 31-8220 significantly reduced the interaction of moesin, but not ezrin, with L-selectin. Alanine mutations of membrane-proximal basic amino acid residues in the cytoplasmic domain of L-selectin identified arginine 357 as a critical residue for both ezrin and moesin interaction. Finally, BIACore affinity analysis confirmed that N-terminal moesin interacts specifically with L-selectin cytoplasmic tail, with relatively high affinity (K_d apprxeq40 nM). Based on these findings, although moesin and ezrin bind to a similar region of the cytoplasmic tail of L-selectin, moesin binding is dependent on PKC activation, which suggests that ezrin and moesin are regulated differently in lymphocytes.

L25 ANSWER 10 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
 ACCESSION NUMBER: 2002:138813 BIOSIS
 DOCUMENT NUMBER: PREV200200138813
 TITLE: Online size-exclusion high-performance liquid chromatography light scattering and differential refractometry methods to determine degree of polymer conjugation to proteins and protein-protein or protein-ligand association states.
 AUTHOR(S): Kendrick, Brent S. [Reprint author]; Kerwin, Bruce A.; Chang, Byeong S.; Philo, John S.
 CORPORATE SOURCE: Amgen, Inc., 4000 Nelson Road, MS AC-3A, Longmont, CO, 80503, USA
 SOURCE: kendrick@amgen.com
 Analytical Biochemistry, (December 15, 2001) Vol. 299, No. 2, pp. 136-146. print.
 CODEN: ANBCA2. ISSN: 0003-2697.
 DOCUMENT TYPE: Article
 LANGUAGE: English
 ENTRY DATE: Entered STN: 6 Feb 2002
 Last Updated on STN: 26 Feb 2002
 AB Characterizing the solution structure of protein-polymer conjugates and protein-ligand interactions is important in fields such as biotechnology and biochemistry. Size-exclusion high-performance liquid chromatography with online classical light scattering (LS), refractive index (RI), and UV detection offers a powerful tool in such characterization. Novel methods are presented utilizing LS, RI, and UV signals to rapidly determine the degree of conjugation and the molecular mass of the protein conjugate. Baseline resolution of the chromatographic peaks is not required; peaks need only be sufficiently separated to represent relatively pure fractions. An improved technique for determining the polypeptide-only mass of protein conjugates is also described. These techniques are applied to determining the degree of erythropoietin glycosylation, the degree of polyethylene glycol conjugation to RNase A and brain-derived neurotrophic factor, and the solution association states of these molecules. Calibration methods for the RI, UV, and LS detectors will also be addressed, as well as online methods to determine protein extinction coefficients and dn/dc values both unconjugated and conjugated protein molecules.

L25 ANSWER 11 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
 ACCESSION NUMBER: 2002:119612 BIOSIS
 DOCUMENT NUMBER: PREV200200119612
 TITLE: Total chemical synthesis of human activin betaA(12-116) and related large-loop polypeptides.
 AUTHOR(S): Keah, Hooi Hong; Allen, Natalie; Clay, Robert; Boysen, Reinhard I.; Warner, Tracy; Hearn, Milton T. W. [Reprint author]
 CORPORATE SOURCE: Center for Bioprocess Technology, Department of Biochemistry and Molecular Biology, Monash University, Clayton, VIC, 3800, Australia
 milton.hearn@med.monash.edu.au
 SOURCE: Biopolymers, (2001) Vol. 60, No. 4, pp. 279-289. print.
 CODEN: BIPMAA. ISSN: 0006-3525.
 DOCUMENT TYPE: Article
 LANGUAGE: English
 ENTRY DATE: Entered STN: 30 Jan 2002
 Last Updated on STN: 26 Feb 2002

AB We report here the synthesis, purification, and characterization of several large polypeptides related to the human activin betaA subunit and their cyclic counterparts. In particular, we describe for the first time the total chemical synthesis of a 105-mer polypeptide, des(1-11) activin betaA, and related large-loop polypeptide, by an optimized solid phase synthetic protocol based on 9-fluorenylmethoxycarbonyl (Fmoc) chemistry. These studies show that automated chemical synthesis utilizing Fmoc-based solid phase synthetic strategies provides a practical alternative to recombinant DNA technology for the production of activin-related subunits, with the opportunity to rapidly provide different analogues and structural variants for subsequent structure-function and associated biophysical investigations.

L25 ANSWER 12 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
 ACCESSION NUMBER: 2001:188939 BIOSIS
 DOCUMENT NUMBER: PREV200100188939
 TITLE: Electrochemical sensor detecting free sulfhydryl groups: Evaluation of milk heat treatment.
 AUTHOR(S): Cosio, M. S.; Mannino, S. [Reprint author]; Buratti, S.
 CORPORATE SOURCE: Department of Food Science and Technology, University of Milan, Via Celoria 2, 20133, Milan, Italy
 saverio.mannino@unimiv.it
 SOURCE: Journal of Dairy Science, (September, 2000) Vol. 83, No. 9, pp. 1933-1938. print.
 CODEN: JDSCAE. ISSN: 0022-0302.

DOCUMENT TYPE: Article

LANGUAGE: English

ENTRY DATE: Entered STN: 20 Apr 2001

Last Updated on STN: 18 Feb 2002

AB We describe a new and rapid method for the evaluation of reactive sulfhydryl groups in whey proteins obtained after precipitation of casein by acetic acid at pH 4.6. The procedure is based on the use of a wire tungsten electrode operating at -0.2 V versus saturated calomel electrode in flow injection analysis. The method was applied to raw milks and to commercial pasteurized and UHT milks. Results showed that the tungsten electrode constituted a robust amperometric sensor that could be used to differentiate milks that underwent different heat treatments. The decrease of thiol content in the whey proteins from samples was in agreement with the whey protein content found by HPLC. The procedure is suitable for on-line quality control of heat-treated milks.

L25 ANSWER 13 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
 ACCESSION NUMBER: 2000:160084 BIOSIS
 DOCUMENT NUMBER: PREV200000160084
 TITLE: Rapid simultaneous detection of two orchid viruses using LC- and/or MALDI-mass spectrometry.
 AUTHOR(S): Tan, Stella Wei-Ling; Wong, Sek-Man [Reprint author]; Kini, R. Manjunatha
 CORPORATE SOURCE: Department of Biological Sciences, National University of Singapore, Kent Ridge, 117543, Singapore
 SOURCE: Journal of Virological Methods, (March, 2000) Vol. 85, No. 1-2, pp. 93-99. print.
 CODEN: JVMEHD. ISSN: 0166-0934.

DOCUMENT TYPE: Article

LANGUAGE: English

ENTRY DATE: Entered STN: 26 Apr 2000

Last Updated on STN: 4 Jan 2002

AB Liquid chromatography/mass spectrometry (LC/MS) and matrix-assisted laser desorption-ionization (MALDI) mass spectrometry are

capable of providing molecular mass information on biological samples with high speed, accuracy and sensitivity. With mass spectrometry, identifying a virus based on the molecular weight of its coat protein is relatively simple and accurate. The technique can be applied to all viruses with known coat **protein molecular** weights. Using the LC/MS and/or MALDI, this paper describes **rapid** simultaneous **detection** of the two most prevalent orchid viruses, namely cymbidium mosaic potexvirus (CymMV) and odontoglossum ringspot tobamovirus (ORSV). The coat **protein molecular** weights of CymMV and ORSV were **detected** accurately using an extract from 1 g of virus-infected Oncidium orchid flower. Because LC/MS and MALDI allow automated analyses of multiple samples with simple preparation steps, both techniques are ideal for **rapid** identification of viruses from a large number of samples. This is the first report on the application of LC/MS and/or MALDI for simultaneous **detection** of two plant viruses from an infected plant extract.

L25 ANSWER 14 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
 ACCESSION NUMBER: 1998:372071 BIOSIS
 DOCUMENT NUMBER: PREV199800372071
 TITLE: Aerolysin induces G-protein activation and Ca²⁺ release from **intracellular** stores in human granulocytes.
 AUTHOR(S): Krause, Karl-Heinz; Fivaz, Marc; Monod, Antoinette; Van Der Goot, F. Gisou [Reprint author]
 CORPORATE SOURCE: Dep. Biochemistry, Univ. Geneva, 30 quai E. Ansermet, 1211 Geneva 4, Switzerland
 SOURCE: Journal of Biological Chemistry, (July 17, 1998) Vol. 273, No. 29, pp. 18122-18129. print.
 CODEN: JBCHA3. ISSN: 0021-9258.
 DOCUMENT TYPE: Article
 LANGUAGE: English
 ENTRY DATE: Entered STN: 27 Aug 1998
 Last Updated on STN: 21 Oct 1998

AB Aerolysin is a **pore**-forming toxin that plays a key role in the pathogenesis of *Aeromonas hydrophila* infections. In this study, we have analyzed the effect of aerolysin on human granulocytes (HL-60 **cells**). Proaerolysin could bind to these **cells**, was processed into active aerolysin, and led to membrane depolarization, indicating that granulocytes are potential targets for this toxin. Fura-2 measurements were used to analyze the effect of aerolysin on cytosolic (Ca²⁺) homeostasis. As expected for a **pore**-forming toxin, aerolysin addition led to Ca²⁺ influx across the plasma membrane. In addition, the toxin triggered Ca²⁺ release from agonist and thapsigargin-sensitive **intracellular** Ca²⁺ stores. This Ca²⁺ release was independent of the aerolysin-induced Ca²⁺ influx and occurred in two kinetically distinct phases: an initial **rapid** and transient phase and a second, more sustained, phase. The first, but not the second phase was sensitive to pertussis toxin. Activation of pertussis toxin-sensitive G-proteins appeared to be a consequence of **pore** formation, rather than receptor activation through aerolysin-binding, as it: (i) was not observed with a binding competent, insertion-incompetent aerolysin mutant, (ii) had a marked lag time, and (iii) was also observed in response to other bacterial **pore**-forming toxins (staphylococcal alpha-toxin, streptolysin O) which are thought to bind to different receptors. G-protein activation through **pore**-forming toxins stimulated **cellular** functions, as evidenced by pertussis toxin-sensitive chemotaxis. Our results demonstrate that granulocytes are potential target **cells** for aerolysin and that in these **cells**, Ca²⁺ signaling in response to a **pore**-forming toxin involves **G-protein** independent

cell activation and Ca²⁺ release from intracellular stores.

L25 ANSWER 15 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1997:37726 BIOSIS

DOCUMENT NUMBER: PREV199799329714

TITLE: A procedure for protein elution from reverse-stained polyacrylamide gels applicable at the low picomole level: An alternative route to the preparation of low abundance proteins for **microanalysis**.

AUTHOR(S): Castellano-Serra, Lila R. [Reprint author];

CORPORATE SOURCE: Fernandez-Patron, Carlos; Hardy, Eugenio; Huerta, Vivian Div. Physical Chem., Center Genetics Engineering Biotechnol., P.O. Box 6162, La Habana 10600, Cuba

SOURCE: Electrophoresis, (1996) Vol. 17, No. 10, pp. 1564-1572.
CODEN: ELCTDN. ISSN: 0173-0835.

DOCUMENT TYPE: Article

LANGUAGE: English

ENTRY DATE: Entered STN: 28 Jan 1997

Last Updated on STN: 28 Jan 1997

AB We developed a technique that allows **rapid** protein elution from polyacrylamide gel bands at room temperature into a detergent-free buffer (elution time 2 X 10 min, total working time about 30 min) with high yields (90-98%) even at a low picomole level (1 picomole per band). Its efficacy relies on the combination of protein **detection** by reverse staining with the enhancement of protein diffusion after gel crushing. **Detection** is accomplished by gel incubation in an imidazole solution, followed by incubation in a zinc salt solution to develop a negative stain pattern. Proteins are eluted by zinc complexation in Laemmli electrophoresis buffer (Tris + glycine), from which sodium dodecyl sulfate is omitted to allow direct subsequent **microanalysis**, e.g. high performance liquid **chromatography** (HPLC) and automatic sequencing. A variety of proteins were eluted efficiently (with no apparent restriction due to their intrinsic properties) as quantified with radioiodinated total E. coli proteins. Yields were independent of acrylamide concentration, **protein molecular mass** (from 10 to 100 kDa) and the amount (from 1 to 100 picomole) of protein in the band. This protocol was derived from a quantitative evaluation of the effect of protein staining and of sample reduction prior to electrophoresis on elution yields. For N-terminal sequencing, the protein eluate was automatically loaded on a polyvinylidene difluoride (PVDF) membrane with conventional HPLC **equipment**; both loading and membrane clean-up were monitored at 206 nm. By simultaneously processing several analytical bands, the procedure allowed trace enrichment of a natural scarce protein that was N-terminal sequenced.

L25 ANSWER 16 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN

ACCESSION NUMBER: 1983:333098 BIOSIS

DOCUMENT NUMBER: PREV198376090590; BA76:90590

TITLE: PURIFICATION AND CHARACTERIZATION OF A NEW SODIUM TRANSPORT DECARBOXYLASE METHYLMALONYL COENZYME A DECARBOXYLASE EC-4.1.1.41 FROM VEILLONELLA-ALCALESCENS.

AUTHOR(S): HILPERT W [Reprint author]; DIMROTH P

CORPORATE SOURCE: INST PHYSIOL CHEMIE, TECHNISCHEN UNIV MUENCHEN, BIEDERSTEINER STR 29, D-8000 MUENCHEN 40, FRG

SOURCE: European Journal of Biochemistry, (1983) Vol. 132, No. 3, pp. 579-588.

CODEN: EJBCAI. ISSN: 0014-2956.

DOCUMENT TYPE: Article

FILE SEGMENT: BA
 LANGUAGE: ENGLISH

AB Upon resolution of the particulate **cell** fraction of *V. alcalescens* by gel **chromatography**, membranes and ribosomes were clearly resolved. Methylmalonyl-CoA decarboxylase was bound to the membranes and not to ribosomes as reported earlier. Membrane vesicles **containing** methylmalonyl-CoA decarboxylase were prepared by disrupting *V. alcalescens* **cells** with a French pressure chamber. About 64% of the decarboxylase was oriented in these vesicles with the substrate binding site facing to the outside. The vesicles performed a **rapid** accumulation of Na⁺ ions in response to the decarboxylation of methylmalonyl-CoA. Decarboxylation and transport were highly uncoupled. The efficiency of the transport was considerably increased if methylmalonyl-CoA decarboxylation was retarded by using a low temperature or by slowly generating the substrate enzymically from propionyl-CoA. Under optimized conditions Na⁺ was concentrated inside the inverted vesicles 8 times higher than in the incubation medium. Methylmalonyl-CoA decarboxylase was solubilized from the membranes with Triton X-100 and purified about 20-fold by affinity **chromatography** on monomeric avidin-Sepharose columns. The decarboxylase was specifically activated by Na⁺ ions (apparent Km ≈ 0.6 mM). Whereas (S)-methylmalonyl-CoA was the superior substrate (apparent Km ≈ 7 μM), malonyl-CoA was also decarboxylated (apparent Km ≈ 35 μM). The decarboxylation of methylmalonyl-CoA yielded CO₂ and not HCO₃⁻ as the primary reaction **product**. **Analysis** of the purified enzyme by dodecylsulfate gel electrophoresis indicated the presence of 4 different polypeptides α, β, γ, δ, with MW 60,000, 33,000, 18,500 and 14,000. The latter of these polypeptides was clearly visible only after Ag-staining, but not after staining with Coomassie brilliant blue. A low MW polypeptide with similar staining properties was also found in oxaloacetate decarboxylase. Methylmalonyl-CoA decarboxylase **contained** about 1 mol covalently bound biotin/125,500 g protein which was localized exclusively in the γ-subunit. This subunit represents the biotin carboxyl carrier protein of methylmalonyl-CoA decarboxylase. A new very sensitive **method** for the **detection** of biotin-containing proteins is described.

L25 ANSWER 17 OF 24 WPIDS COPYRIGHT 2003 THOMSON DERWENT on STN
 ACCESSION NUMBER: 2003-663395 [62] WPIDS
 DOC. NO. CPI: C2003-180197
 TITLE: RNA-DNA ligation **products** and nucleic acid constructs, useful e.g. in screening nucleic acids or proteins, producing protein-DNA or RNA complexes, and **cell-free** translation systems to express target peptides.
 DERWENT CLASS: B04 D16
 INVENTOR(S): NEMOTO, N; SASAKI, T; SHIRATORI, M
 PATENT ASSIGNEE(S): (MITU) MITSUBISHI CHEM CORP
 COUNTRY COUNT: 101
 PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
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WO 2003062417 A1 20030731 (200362)* JA 159
RW: AT BE BG CH CY CZ DE DK EA EE ES FI FR GB GH GM GR HU IE IT KE LS LU MC MW MZ NL OA PT SD SE SI SK SL SZ TR TZ UG ZM ZW
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CO CR CU CZ DE DK DM DZ EC EE ES FI GB GD GE GH GM HR HU ID IL IN IS KE KG KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ OM PH PL PT RO RU SC SD SE SG SK SL TJ TM TN TR TT TZ UA UG US UZ VC VN YU ZA ZM ZW

JP 2003299489 A	20031021 (200370)	15
JP 2003313198 A	20031106 (200375)	11

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2003062417 A1		WO 2003-JP544	20030122
JP 2003299489 A		JP 2003-30680	20030207
JP 2003313198 A		JP 2003-42428	20030220

PRIORITY APPLN. INFO: JP 2002-211405 20020719; JP 2002-12820
 20020122; JP 2002-31779 20020208; JP
 2002-44955 20020221

AN 2003-663395 [62] WPIDS

AB WO2003062417 A UPAB: 20030928

NOVELTY - Producing an RNA-DNA ligation **product** comprising annealing a single-stranded RNA with a single-stranded DNA which have sequences complementary to each other, and treating the annealing **product** with an RNA ligase for ligation of 3'-terminal of such single-stranded RNA with 5'-terminal of the single-stranded DNA or its derivative, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for:

(1) producing an RNA-DNA ligation **product** by:

(a) annealing a single-stranded RNA **containing** an annealing sequence from 5'-3' direction at the 3'-terminal with coding sequence of a protein and branch sequence, with a single-stranded DNA **containing** annealing sequence from 3'-5' direction, complementary sequence and branch sequence, or its 5'-terminal derivative; and

(b) ligating the annealing **product** as already specified;

(2) RNA-DNA ligation **products** produced by the

method of (1);

(3) producing a DNA ligation **product** by performing reverse transcription with the obtained RNA-DNA ligation **product**;

(4) producing an RNA-protein complex **containing** such RNA and its encoded protein by introducing the obtained RNA-DNA ligation **product** into a protein translation system for the RNA to translate the protein;

(5) RNA-protein complexes thus produced;

(6) producing a nucleic acid-protein complex made from a DNA and its encoded protein by performing reverse transcription with the already-obtained RNA-protein complex;

(7) nucleic acid complexes produced by the **method** of (6);

(8) a nucleic acid construct produced from the single-stranded RNA and DNA, a primer sequence for reverse transcription and a nucleic acid derivative with a space branched out for bonding and producing a complex of the RNA and its encoded protein;

(9) producing the nucleic acid construct in which spacer branches of the nucleic acid derivative is for bonding, and the single-stranded RNA without being annealed in the 5'-terminal side in the construct forms an inter-complementary double-stranded sequence via a loop region that is bonded to a substance with affinity;

(10) producing an RNA-DNA ligation **product** by annealing the nucleic acid construct with a single-stranded RNA before ligation;

(11) RNA-DNA ligation **products** produced from the nucleic acid construct;

(12) chips made by immobilizing RNA-DNA ligation **products** onto a support;

(13) producing a DNA ligation **product** by performing reverse

transcription with the nucleic acid construct-based RNA-DNA ligation **product**;

(14) DNA ligation **products** produced by the **method** of (13);

(15) chips obtained by immobilizing the DNA ligation **products** onto a support;

(16) producing an RNA-protein complex by introducing a RNA-DNA ligation **product** into a protein translation system for the protein-encoded RNA to start the protein translation;

(17) RNA-protein complexes obtained by the **method** of (16);

(18) chips produced by immobilizing the RNA-protein complexes onto a support;

(19) producing a nucleic acid-protein complex by performing reverse transcription with the RNA-protein complex that **contains** a protein-encoded nucleic acid;

(20) nucleic acid-protein complexes produced by the **method** of (19);

(21) chips obtained by immobilizing the nucleic acid-protein complexes onto a support;

(22) selecting a nucleic acid and/or a protein by using any of the ligation **products** of a target mRNA and its encoded protein through interaction and nucleic acid amplification optionally repeatedly and with mutation;

(23) **detecting** substances interacting with a protein using the ligation **products**; and

(24) puromycin derivatives of formula (I) or their salts;

(25) puromycin derivative-immobilized support obtained by bonding such derivative to the support;

(26) deblocking the puromycin derivatives by treatment with peptidase or proteinase;

(27) producing nucleic acid compounds by using the puromycin derivatives or puromycin derivative-immobilized supports;

(28) nucleic acid compounds containing the puromycin derivatives;

(29) deprotecting puromycin derivatives that contain nucleic acid compounds by using the peptidase or proteinase;

(30) supporter proteins for expressing or presenting target peptides or protein as their fusion proteins, which are spherical proteins made from 30-200 amino acid residues;

(31) supporter proteins containing a 73 amino acid sequence, given in the specification, or one producing a spherical protein based on the sequence of (XXI) but with some amino acids deleted, substituted, added and/or inserted;

(32) nucleic acid or its modifier encoding the fusion protein in which a sequence for the supporter protein is bonded directly or through a linker to that of the target peptide or protein;

(33) fusion proteins produced from the supporter protein and target peptides or proteins;

(34) producing a fusion protein by expressing the nucleic acid or its modifier in a cell-free translation system or live cells;

(35) producing a complex made from the fusion protein-encoded nucleic acid by expressing the fusion protein-encoded mRNA that is bonded to a nucleic acid derivative at its 3'-terminal side, in a cell-free translation system or live cells;

(36) screening functional peptides or proteins by using the fusion proteins in a library for selection of the required biological activity;

(37) producing single-stranded RNAs by using the RNA-DNA ligation products with DNA synthase for reaction in the absence of a primer having not less than 2 different types of single or double-stranded DNAs with inter-complementary common sequences, followed by transcription reaction in the presence of an RNA polymerase to give RNAs and decomposition of the

DNA with a DNA-decomposing enzyme;

(38) RNAs produced by the method of (37);

(39) a process for producing proteins by expressing the thus obtained RNA in a cell-free translation system or live cells; and

(40) producing complexes of proteins and their encoded nucleic acids by modifying the 3'-terminal of such obtained RNAs to give nucleic acids for expression in a cell-free translation system or live cells.

R1 = H or hydroxyl-protecting group;

R2 = H or reactive group; and

X = amino acid residue or peptide, provided that when the carboxyl group in X is linked to amino group in the puromycin via an amide bond, alpha -amino group of such amino acid residue or functional group of the side-chain is suitably protected.

USE - The ligation products and nucleic acid constructs are useful in screening nucleic acids or proteins, producing protein-DNA or RNA complexes, cell-free translation systems to express target peptides or novel proteins by evolution molecular engineering, for in vitro synthesis of virus genomes, and post-genomic protein-interaction studies.

ADVANTAGE - The RNA-DNA ligation products can be produced efficiently and quickly by this method. The method can also stabilize the protein-DNA complexes and for the effective and selective detection and functional analysis of biologically-active proteins and/or nucleic acids as well.

Dwg.0/25

L25 ANSWER 18 OF 24 WPIDS COPYRIGHT 2003 THOMSON DERWENT on STN
 ACCESSION NUMBER: 2003-256367 [25] WPIDS
 DOC. NO. CPI: C2003-066416
 TITLE: Reagent for mass spectrometric **analysis** of
 proteins, comprises tag molecule having reactive site for
 stably associating with protein, isotope label and pH
 sensitive anchoring site for anchoring tag to solid
 phase.
 DERWENT CLASS: B04 D16
 INVENTOR(S): GARTNER, C A; GERBER, S A; GYGI, S P
 PATENT ASSIGNEE(S): (HARD) HARVARD COLLEGE
 COUNTRY COUNT: 100
 PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2003008547	A2	20030130	(200325)*	EN	49
RW:	AT BE BG CH CY CZ DE DK EA EE ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SK SL SZ TR TZ UG ZM ZW				
W:	AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CO CR CU CZ DE DK DM DZ EC EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ OM PH PL PT RO RU SD SE SG SI SK SL TJ TM TN TR TT TZ UA UG US UZ VN YU ZA ZM ZW				

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2003008547	A2	WO 2002-US22598	20020716

PRIORITY APPLN. INFO: US 2001-305808P 20010716

AN 2003-256367 [25] WPIDS

AB WO2003008547 A UPAB: 20030416

NOVELTY - A reagent (I) for mass spectrometric **analysis** of

proteins, comprising a tag molecule (TM), where the tag molecule has a reactive site for stably associating with a protein, an isotope label, and a pH sensitive anchoring site for covalently anchoring the tag molecule to a solid phase, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for:

(1) a **composition** (C1) comprising a pair of TM, where each member of the pair is identical except for the mass of the isotope attached to it;

(2) a **composition** (C2) comprising a reagent for mass spectrometric **analysis** of proteins comprising a first and second tag molecule, where the first tag molecule comprises a reactive site for stably associating with a protein, an isotope label, and a pH sensitive anchoring site for anchoring the tag molecule to a solid phase and the second tag molecule is identical to the first tag molecule but does not comprises an isotope label;

(3) a **kit** (K1) comprising (I), (C1) or (C2) and one or more of a reagent selected from an activating agent for providing active groups on a protein which bind to the reactive site of the tag molecule, a solid phase, one or more agents for lysing a **cell**, a pH altering agent, one or more proteases, one or more **cell** samples or their fractions; and

(4) a **kit** (K2) comprising several tagged **peptide molecules**, each tagged **peptide molecule** comprising a peptide and a tag molecule stably associated with the protein, the tag molecule further comprising an isotope label and a pH sensitive anchoring site for anchoring the tag molecule to a solid phase.

USE - (I) is useful for identifying one or more proteins or protein functions in one or more samples **containing** mixtures of proteins, by reacting a sample with (I) and a solid phase under conditions suitable to form a solid phase-isotope labeled tag molecule-protein complex, digesting the complex with one or more proteases, thus generating solid phase-isotope labeled tag molecule-peptide complexes and untagged peptides, purifying the solid phase-isotope labeled tag molecule-peptide complexes, exposing the solid phase-isotope labeled tag molecule-peptide complexes to a pH which disrupts associations between the anchoring site of the tag molecule and the solid phase, thus releasing a tagged peptide from the solid phase, determining the mass of the tagged peptide, correlating the mass to the identity and/or activity of a protein. The mass-to-charge ratio of the tagged peptide is determined. The **method** for the involves subjecting a sample comprising one or more tagged peptides to a separation step. The separation step comprises liquid **chromatography**. The **method** involves subjecting one or more tagged peptides to mass spectrometric (MS_n) **analysis**, and further involves reacting a second sample with a second reagent comprising an identical molecular tag as the first reagent but which is differentially labeled and combining the two samples prior to protease digestion and generating a combined sample comprising at least one pair of tagged peptides, each member of the pair comprising identical peptides but differing in mass, determining the ratio of members of at least one tagged peptide pair in the combined sample, generating mass spectra comprising at least one signal doublet for each peptide in the sample, the signal doublet comprising a first signal and a second signal shifted a number of known units for the first signal, where the known units represent the difference in molecular weight between the two members of a tagged peptide pair, determining a signal ratio for a given peptide by relating the difference in signal intensity between the first signal and the second signal, and relating mass spectra data from a tagged peptide to an amino acid sequence. The above steps are repeated, either sequentially or simultaneously, until substantially all of the proteins in a sample are **detected** and/or identified. The relative amounts of members of a

tagged peptide pair in the two samples are determined and correlated with the abundance of the protein corresponding to the peptide in the sample. The **method** further involves correlating the relative abundance of the protein with the state of the **cells**, where the correlation is used to diagnose a pathological condition in a patient from whom one of the **cell** samples was obtained, determining the quantity of a protein corresponding to the peptide in the sample, where determining the site of a modification of a protein in one or more samples, by reactive sample proteins with a tag molecule comprising a reactive site which reacts with a modified residue on the protein, and determining the amount of modified protein in the sample. (All claimed.) (I) is useful in quantitative protein expression profiling, for quantitative **analysis** of protein in mixtures of proteins, e.g. to profile the proteome of a **cell** at a particular **cell** state, for the **analysis** of low abundance proteins and classes of proteins with particular physico-chemical properties including poor solubility, large or small size and extreme pH values, to determine sites of protein modifications and thus the abundance of modified proteins in a sample, for qualitative and/or quantitative **analysis** of global protein expression profiles in **cells** and tissues, i.e. **analysis** of proteomes, in clinical and diagnostic analyses to detect the presence, absence, deficiency or excess of a given protein or protein function in a biological fluid (e.g. blood), or in **cells** or tissue.

ADVANTAGE - (I) rapidly and quantitatively analyzes proteins or protein function in mixtures of proteins. Proteins are quantitated directly from **cell** lysates using (I).

Dwg.0/5

L25 ANSWER 19 OF 24 WPIDS COPYRIGHT 2003 THOMSON DERWENT on STN
 ACCESSION NUMBER: 2002-691593 [74] WPIDS
 DOC. NO. NON-CPI: N2002-545608
 DOC. NO. CPI: C2002-195440
 TITLE: **Isolating** biological macromolecules such as nucleic acid molecules or proteins from **cells** or tissues, involves contacting a **filter** with a biological sample comprising biological macromolecules of interest.
 DERWENT CLASS: A89 B04 D16 S03
 INVENTOR(S): SIMMS, D; TRINH, T
 PATENT ASSIGNEE(S): (SIMM-I) SIMMS D; (TRIN-I) TRINH T; (INVI-N) INVITROGEN CORP
 COUNTRY COUNT: 100
 PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2002065125	A1	20020822 (200274)*	EN	42	
RW:	AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SL SZ TR TZ UG ZM ZW				
W:	AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CO CR CU CZ DE DK DM DZ EC EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ OM PH PL PT RO RU SD SE SG SI SK SL TJ TM TN TR TT TZ UA UG UZ VN YU ZA ZM ZW				
US 2002127587	A1	20020912 (200274)			

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
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WO 2002065125 A1	WO 2002-US4185	20020213
US 2002127587 A1 Provisional	US 2001-268027P	20010213
	US 2002-73260	20020213

PRIORITY APPLN. INFO: US 2001-268027P 20010213; US 2002-73260
20020213

AN 2002-691593 [74] WPIDS

AB WO 200265125 A UPAB: 20021118

NOVELTY - **Isolation** (M) of biological macromolecules, involves contacting at least one **filter** with a biological sample comprising the biological macromolecules of interest, where the **pore** size of the **filter** increases in the direction of sample flow.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for:

(1) an **apparatus** (I) for use in **isolating** biological macromolecules, comprising one or more **filters**, where the **pore** size of the **filters** increases in the direction of sample flow;

(2) a **kit** (II) for use in **isolating** a nucleic acid molecule or a population of nucleic acid molecules, comprising (I); and

(3) a device (III) for performing (M), comprising a cylindrical hollow body having inlet and an outlet, where the cylindrical hollow body comprises a **filter** comprising increasing **pore** sizes as seen in the direction of outlet, the **filter** having a **pore** size ranging from 0.1-500 micro m, where the total thickness of the **filter** bed is from 0.1-10 micro m.

USE - (M), (I), (II) or (III) are useful for **isolating** biological macromolecules such as nucleic acid molecules (selected from RNA, mRNA and DNA molecules, where the DNA molecules are vectors or plasmids), where the nucleic acid is plasmid DNA or genomic DNA having a size of from 1-50 kb (kilo base pairs), or **protein molecules** (claimed). The nucleic acid molecules **isolated** by (M) are useful for amplifying and sequencing nucleic acid molecules, and for the manufacture of various materials (such as hybridization probes, therapeutic proteins, gene therapy vehicles and compositions, and molecular weight markers) in industrial processes.

ADVANTAGE - (M) or (II) enables **rapid isolation** of nucleic acid molecules from bacterial **cells**.

Dwg.0/8

L25 ANSWER 20 OF 24 WPIDS COPYRIGHT 2003 THOMSON DERWENT on STN

ACCESSION NUMBER: 2002-257280 [30] WPIDS

DOC. NO. CPI: C2002-076534

TITLE: **Isolating protein and peptide**

molecules from bacterial, fungal, animal, yeast or plant **cells**, comprises using **lysis** /**filter** matrix.

DERWENT CLASS: A89 B04 C06 D16 D25

INVENTOR(S): BLAKESLEY, R. W.; CLAUSEN, P.; FLYNN, B.

PATENT ASSIGNEE(S): (INVI-N) INVITROGEN CORP

COUNTRY COUNT: 97

PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
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WO 2002006456 A1	20020124	(200230)*	EN	83
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RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ

NL OA PT SD SE SL SZ TR TZ UG ZW
 W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CO CR CU CZ DE DK
 DM DZ EC EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR
 KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ PL PT RO RU
 SD SE SG SI SK SL TJ TM TR TT TZ UA UG UZ VN YU ZA ZW
 US 2002012982 A1 20020131 (200230)
 AU 2001073432 A 20020130 (200236)
 EP 1301591 A1 20030416 (200328) EN
 R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT
 RO SE SI TR

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2002006456 A1		WO 2001-US22080	20010713
US 2002012982 A1	Provisional	US 2000-218081P	20000713
	Provisional	US 2001-274630P	20010312
		US 2001-903864	20010713
AU 2001073432 A		AU 2001-73432	20010713
EP 1301591 A1		EP 2001-952705	20010713
		WO 2001-US22080	20010713

FILING DETAILS:

PATENT NO	KIND	PATENT NO
AU 2001073432 A	Based on	WO 2002006456
EP 1301591 A1	Based on	WO 2002006456

PRIORITY APPLN. INFO: US 2001-274630P 20010312; US 2000-218081P 20000713; US 2001-903864 20010713

AN 2002-257280 [30] WPIDS

AB WO 200206456 A UPAB: 20020513

NOVELTY - **Isolating** (I) a **protein molecule** or population of protein or **peptide molecules**, comprising contacting one or more **cellular sources** of protein or **peptide molecules** with a **pore-containing** matrix (M) which retards flow of high molecular weight (MW) molecules, structures and aggregates but does not retard flow of soluble protein and **peptide molecules**, and separating the molecules from the high MW molecules, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following:

(1) an **isolated** protein or **peptide molecule** (II) produced by (I);

(2) a **composition** (III) for use in **isolating** a protein or **peptide molecule** or their population, comprising one or more **cellular sources** of protein or **peptide molecules**, one or more **pore-containing** (M), and optionally a compound or **composition** that lyses/disrupts/permeabilizes the **cellular source**;

(3) an **apparatus** (IV) for extracting and **isolating** protein or **peptide molecules**, comprising a housing and one or more **pore-containing** (M), and a

composition chosen from **chromatographic** resins (CR) that bind proteins or peptides, CR that bind impurities, CR having bound to protein modifying reagents, CR bound to enzymes, nucleic acids, enzyme substrates and **filters**, and **compositions** capable of being used for **detecting** or quantifying the amount of protein or

nucleic acid present in the sample; and

(4) a kit for use in isolating a protein or peptide molecule or their population, comprising (IV).

USE - (I) is useful for isolating protein or peptide molecules from any cell or cellular source, including bacterial cells (particularly Escherichia coli), yeast cells (e.g. Saccharomyces), fungal, animal cell, a cell infected by a virus, or a plant cell (claimed). The animal cells include insect cells and mammalian cells such as human cells, Chinese Hamster Ovary cells, Bowes melanoma cells and HepG2 cells. (I) is well suited for isolation of soluble proteins and peptides expressed from a cDNA expression library or recombinant proteins and peptides expressed from plasmids in a prokaryotic or eukaryotic host. Also suitable for use as sources of protein and peptide molecules are mammalian tissues or cells derived from brain, kidney, liver, pancreas, blood, bone marrow, muscle, skin, connective tissue, or from embryo or fetus. These cells, tissues and organs may be normal, primary, transformed or those involved in infectious diseases caused by bacteria, fungi, yeast, virus (including AIDS) or parasites, in genetic or biochemical pathologies e.g. cystic fibrosis, hemophilia, Alzheimer's disease, schizophrenia, muscular dystrophy or multiple sclerosis, or in cancers or cancerous process. The isolated proteins and peptide molecules are useful in the manufacture of various materials in industrial processes, pharmaceuticals (enzymatic catalysis of pharmaceutical precursors), protein or peptide molecular weight standards, and modification of proteins or peptides, DNA, lipids or carbohydrates by enzymatic catalysis. Additionally, libraries of expressed protein and peptide molecules are screened in a high throughput format using a multiwell plate for the presence of a desired characteristic or activity.

ADVANTAGE - The method allows quick isolation and/or analysis of proteins and peptides from numerous sources.

DESCRIPTION OF DRAWING(S) - The figure shows the apparatus for isolating protein or peptide molecules.

Tube containing porous matrix material 1

Sample application section 3

Sample elution section 4

Peptide binding resin and/or contaminant binding resin 5

Dwg.1/14

L25 ANSWER 21 OF 24	WPIDS COPYRIGHT 2003 THOMSON DERWENT on STN
ACCESSION NUMBER:	2002-010942 [01] WPIDS
CROSS REFERENCE:	2002-097342 [13]; 2002-499091 [53]; 2002-626184 [67]; 2003-596266 [56]; 2003-764866 [72]
DOC. NO. NON-CPI:	N2002-009086
DOC. NO. CPI:	C2002-002761
TITLE:	Screening for bioactivity of candidate compound towards target proteins in mixture, useful for generating large number of drug molecules, comprises combining probe with mixture and sequestering proteins conjugated to probe.
DERWENT CLASS:	B04 D16 S03 T01
INVENTOR(S):	ADAM, G; CRAVATT, B F; LOVATO, M; PATRICELLI, M; SORENSEN, E
PATENT ASSIGNEE(S):	(SCRI) SCRIPPS RES INST
COUNTRY COUNT:	94
PATENT INFORMATION:	

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2001077668	A2	20011018 (200201)*	EN	118	
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ					
NL OA PT SD SE SL SZ TR TZ UG ZW					
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CR CU CZ DE DK DM					
DZ EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC					
LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ PL PT RO RU SD SE					
SG SI SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW					
AU 2001024349	A	20011023 (200213)			

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2001077668	A2	WO 2000-US34167	20001215
AU 2001024349	A	AU 2001-24349	20001215

FILING DETAILS:

PATENT NO	KIND	PATENT NO
AU 2001024349 A	Based on	WO 2001077668

PRIORITY APPLN. INFO: US 2000-222532P 20000802; US 2000-195954P
20000410; US 2000-212891P 20000620

AN 2002-010942 [01] WPIDS
CR 2002-097342 [13]; 2002-499091 [53]; 2002-626184 [67]; 2003-596266 [56];
2003-764866 [72]

AB WO 2001077668 A UPAB: 20031107
NOVELTY - Screening for the bioactivity of candidate compound toward a group of related target proteins in proteomic mixture of proteins from **cell** comprising:

- (a) combining a probe with an untreated portion and a portion inactivated with a non-covalent agent;
- (b) sequestering proteins conjugated with the probe;
- (c) determining the proteins that are sequestered; and
- (d) comparing amount of the proteins sequestered, is new.

DETAILED DESCRIPTION - Screening (M1) for the bioactivity of a candidate compound toward a group of related target proteins in a proteomic mixture of proteins from a **cell**, by employing at least one probe comprising:

- (a) combining at least one probe with an untreated portion and with a portion inactivated with a non-covalent agent, of the mixture under conditions for reaction with the target proteins;
- (b) sequestering proteins conjugated with the probe from each of the mixtures;
- (c) determining the proteins that are sequestered; and
- (d) comparing the amount of each of the proteins sequestered from the untreated portion and the inactivated portion as indicative of the bioactivity of the candidate compound with the target proteins. The probe comprises a reactive functionality group specific for the group of target proteins and a ligand.

INDEPENDENT CLAIMS are also included for the following:

(1) screening for the bioactivity of a candidate compound toward a group of related target enzymes in a proteomic mixture of proteins from a **cell** employing at least one probe of formula R asterisk (F-L)-X

(I) comprising M1;

(2) determining in a proteomic mixture (A) the presence of active target members (B) comprising a group of related proteins involving:

(a) combining (A) in wild-type form with a probe;
 (b) combining (A) after non-specific deactivation with the probe; and
 (c) determining the presence of (B) conjugated with the probe in (A)
 in active and inactive form, where the probe comprises a reactive
 functionality specific for the active site when active, under conditions
 for conjugation of the probe to (B) and when the probe conjugated to (B)
 in (A) in active form and in less amount in inactive form, the presence of
 (B) is determined;

(3) determining in a plurality of proteomic mixtures the presence of
 active target members of a group of related proteins which have a common
 functionality for conjugation at an active site comprising:

(a) combining the mixtures in wild type form with a probe
containing a reactive functionality specific for the active site;
 (b) determining the presence of target members conjugated with the
 probe; and
 (c) analyzing for the presence of target members conjugated with the
 probe using simultaneous individual capillary electrokinetic
analysis or capillary high performance liquid
chromatography (HPLC), where when the target members are
 conjugated to target members, the presence of active target members is
 determined;

(4) determining in a proteomic mixture the presence of active target
 members of a group of related enzymes which have common functionality for
 conjugation at an active site comprising:

(a) combining the mixture in wild type form with a probe
containing a reactive functionality specific for the active site;
 (b) combining the mixture after non-specific deactivation with the
 probe;
 (c) determining the presence of target members conjugated with the
 probe in the proteomic mixtures in active and inactive form, where the
 probe is conjugated to at least one target member in the mixture in active
 form and in lesser amount in inactive form, the presence of active members
 is determined;

(5) a system for identifying active target proteins in a related
 group of proteins in a sample, using at least one activity-based probe
 (ABP) binding to several members of the proteins comprising:

(a) a sample **containing** at least one of the target protein;
 (b) ABP of formula R asterisk (Q-L)-X (II); and
 (c) a programmed data processor for receiving and transmitting values
 comprising a program for evaluating results from the combining of ABP and
 sample resulting in formation of conjugates with active target proteins
 present to determine the presence of active target proteins and providing
 a profile of the binding;

(6) a system for determining the status of a biological system in
 relation to the presence of members of at least one related group of
 active proteins, by employing the results from combining (I) and a sample
 suspected of **containing** at least one target protein, to produce
 conjugates of (I) with the target proteins in varying amounts in relation
 to the amount of each of the active target proteins.

X = a ligand for binding to a reciprocal receptor or a chemically
 reactive functionality for reacting with a reciprocal functionality for
 adding a ligand;

L = a linking group, which is the same in each of the members of a
 library;

Q = a functional group reactive at an active site of a target
 protein, and is the same reactive functionality in each of the members of
 the library (preferably a sulfonyl group, fluorophosphonyl or
 fluorophosphoryl group); and

R asterisk = H or a moiety of less than 1 kDa providing specific
 affinity for the target protein;

asterisk = intends that R is a part of F or L.
 F = functional group reactive at an active site of a target enzyme and is the same reactive functionality in each of the members of the library.

USE - For screening for the bioactivity of a candidate compound towards a group of related target proteins; e.g. for determining the status of a biological system in relation to the presence of the active protein; such as an infectious disease, a response to a therapeutic agent or a response to a candidate drug (claimed). The method is also useful for rapidly generating and developing large numbers of drug candidate molecules or for randomly generating a large number of drug candidates and later optimizing those candidates that show the most medicinal promise; for systematically synthesizing a large number of molecules that may vary greatly in their chemical structure or composition or that may vary in minor aspects of their chemical structure or composition. The screened compounds can be used to indicate the presence of a particular disease in a human or animal, the compounds can stimulate or inhibit the activity of bacteria, viruses, fungi or other infectious agent and/or modulate the effect of a disease by preventing or decreasing the severity of disease or curing a disease such as cancer, diabetes, atherosclerosis, high blood pressure, Parkinson's disease and other disease states.

ADVANTAGE - The method easily identifies the biological target molecule for lead compounds, all with varying ability to block cell division. The method shows whether the multiple lead compounds interact with the same or different biological target molecules. The method is simple, takes less time and is economical.

Dwg.0/24

L25 ANSWER 22 OF 24 WPIDS COPYRIGHT 2003 THOMSON DERWENT on STN
 ACCESSION NUMBER: 2001-522222 [57] WPIDS
 DOC. NO. CPI: C2001-155864
 TITLE: Essentially pure daptomycin, useful for treating infections caused by gram-positive bacteria, also **method** for purifying it from fermentation broth.
 DERWENT CLASS: B05 C03 D16
 INVENTOR(S): DECOURCEY, J P; KELLEHER, T J; LAI, J; LYNCH, P D;
 TAGLIANI, A R; ZENONI, M
 PATENT ASSIGNEE(S): (CUBI-N) CUBIST PHARM INC
 COUNTRY COUNT: 95
 PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2001053330	A2	20010726	(200157)*	EN	94
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SL SZ TR TZ UG ZW					
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CR CU CZ DE DK DM DZ EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW					
AU 2001030978	A	20010731	(200171)		
BR 2001007731	A	20021001	(200268)		
NO 2002003476	A	20020920	(200275)		
EP 1252179	A2	20021030	(200279)	EN	
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT RO SE SI TR					
CZ 2002002830	A3	20021113	(200282)		
KR 2002083151	A	20021101	(200319)		
HU 2002003969	A2	20030328	(200333)		
CN 1404487	A	20030319	(200344)		

JP 2003520807 W 20030708 (200347) 118

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2001053330	A2	WO 2001-US1748	20010118
AU 2001030978	A	AU 2001-30978	20010118
BR 2001007731	A	BR 2001-7731	20010118
		WO 2001-US1748	20010118
NO 2002003476	A	WO 2001-US1748	20010118
		NO 2002-3476	20020719
EP 1252179	A2	EP 2001-903121	20010118
		WO 2001-US1748	20010118
CZ 2002002830	A3	WO 2001-US1748	20010118
		CZ 2002-2830	20010118
KR 2002083151	A	KR 2002-709279	20020719
HU 2002003969	A2	WO 2001-US1748	20010118
		HU 2002-3969	20010118
CN 1404487	A	CN 2001-805212	20010118
JP 2003520807	W	JP 2001-553802	20010118
		WO 2001-US1748	20010118

FILING DETAILS:

PATENT NO	KIND	PATENT NO
AU 2001030978	A Based on	WO 2001053330
BR 2001007731	A Based on	WO 2001053330
EP 1252179	A2 Based on	WO 2001053330
CZ 2002002830	A3 Based on	WO 2001053330
HU 2002003969	A2 Based on	WO 2001053330
JP 2003520807	W Based on	WO 2001053330

PRIORITY APPLN. INFO: US 2000-735191 20001128; US 2000-177170P
20000120

AN 2001-522222 [57] WPIDS

AB WO 200153330 A UPAB: 20011005

NOVELTY - Essentially pure daptomycin (I).

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following:

(1) pharmaceutical composition (C1) containing (I);

(2) method for purifying (I);

(3) isolated compounds designated CB-131012, -131011,

-131008, -131006, -130989, -131005, -131009 and -131078;

(4) method of treatment comprising administering the (C);

(5) a lipopeptide micelle comprising a

lipopeptide (Ia) selected from (I), A54145, (I)-related lipopeptides, or an antibiotic A-21978 in which the n-decanoyl side chain is replaced by some other n-8-14C alkanoyl residue;

(6) pharmaceutical composition containing (Ia);

(7) method of treatment comprising administering the (C2);

and

(8) methods for preparing (Ia) and (C2).

ACTIVITY - Antibacterial.

MECHANISM OF ACTION - No details provided.

USE - (I), or lipoprotein micelles

containing it or related antibiotics, are used for treating infections, particularly those caused by gram-positive bacteria (including

strains resistant to conventional antibiotics), e.g. respiratory or urinary tract infections, intra-abdominal sepsis, endocarditis and nephritis and many others.

ADVANTAGE - High purity (I) can be produced on a commercial scale and with reduced solvent content. By fermentation in presence of a low residual content of decanoic acid, **production** of (I) is increased. (I) has a very **rapid** antibacterial action.

Dwg.0/15

L25 ANSWER 23 OF 24 WPIDS COPYRIGHT 2003 THOMSON DERWENT on STN
 ACCESSION NUMBER: 2000-349567 [30] WPIDS
 CROSS REFERENCE: 1998-052326 [05]; 2000-464335 [40]; 2002-654433 [70]
 DOC. NO. CPI: C2000-106241
 TITLE: Identifying, comparing and **detecting** inhibitors of protein-protein interactions within population of host **cells**, involves **detecting** regulation of transcription of nucleic acid sequence by fusion protein interaction.
 DERWENT CLASS: B04 D16
 INVENTOR(S): KALBFLEISCH, T S; KNIGHT, J R; NANDABALAN, K; ROTHBERG, J M; YANG, M
 PATENT ASSIGNEE(S): (CURA-N) CURAGEN CORP
 COUNTRY COUNT: 1
 PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
US 6057101	A	20000502 (200030)*		161	

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
US 6057101	A CIP of	US 1996-663824 US 1997-874825	19960614 19970613

PRIORITY APPLN. INFO: US 1997-874825 19970613; US 1996-663824 19960614

AN 2000-349567 [30] WPIDS
 CR 1998-052326 [05]; 2000-464335 [40]; 2002-654433 [70]
 AB US 6057101 A UPAB: 20021105

NOVELTY - **Detecting** (D) at least 1 protein-protein interaction (PPI) by recombinantly expressing within a population of host **cells**, populations of first and second fusion proteins comprising DNA binding domain (DBD) and transcriptional regulatory domain (TRD) respectively and **detecting** the regulation of transcription of nucleotide sequence of host **cells** operably linked to a promoter driven by DBD, is new.

DETAILED DESCRIPTION - Novel **method** of (D) comprises recombinantly expressing (within a population of host **cells**) a population of fusion proteins (FP1) with a complexity of at least 1000, comprising a first protein sequence and a DBD which is same for all FP1's, and a second population of fusion proteins (FP2) with a complexity of at least 1000, comprising a second protein sequence and a TRD which is same for all FP2's. FP1 and FP2 are coexpressed within host **cells** containing at least 1 nucleotide sequence driven by at least 1 DNA binding site recognized by a DBD, and the interaction between FP1 and FP2 is **detected** by **detecting** the transcription of the nucleotide sequence regulated by the interaction between the 2 proteins.

INDEPENDENT CLAIMS are also included for the following:

(1) **detecting** an inhibitor of PPI comprising incubating a population of **cells** recombinantly expressing FP1 and FP2, in the presence of at least 1 candidate inhibitor molecule in an environment in which the substantial death of **cells** occurs when FP1 and FP2 interact, when increased transcription of FP1 occurs or if the **cells** lack a recombinant nucleic acid encoding FP1 and FP2 and **detecting** those **cells** that survive;

(2) determining at least 1 characteristic or identity of nucleic acids encoding an interacting pair of proteins from a population of **cells containing** nucleic acids encoding different pairs of interacting proteins comprises:

(a) designating each group of **cells containing** nucleic acids as 1 point of multidimensional array in each dimension, uniquely identifying a single group;

(b) pooling all groups along a simple axis to form a number of pooled groups;

(c) amplifying each first and second nucleic acids encoding FP1 and FP2 respectively;

(d) subjecting the nucleic acid for size separation; and

(e) identifying the presence of first and second nucleic acids in the samples of pooled groups from each axis in each dimension and identifying that they code for FP1 and FP2 by indicating their presence at the same intersection in the array;

(3) purified **cells** (PC) of the single yeast strain of mating type a or alpha that is mutant in endogenous URA3 and HIS3 and **containing** functional URA3 and lacZ coding sequences under the control of a promoter **containing** GAL4 binding **cells**;

(4) a **kit** (K) comprising PC in one or more **containers**;

(5) a purified expression vector comprising a first nucleotide sequence encoding a peptide of at least 20 amino acids (aa) fused to a nuclear localization signal operatively linked to the promoter, a yeast **cell** and Escherichia coli **cell** replicating unit, a second nucleotide sequence encoding a yeast selectable marker linked to transcriptional promoter and transcription terminal signal and a third nucleotide sequence encoding an Escherichia coli selectable marker;

(6) **detecting** and recording at least PPI by selecting the positive host **cells** that coexpressed FP1 and FP2 with an increased transcription of at least one nucleotide sequence and updating the first computer-implemented data-store with an information in digital form characterized for selected host **cell** and for FP1 and FP2;

(7) a computer-implemented **method** for storing and analyzing at least a pair-wise interaction between protein sequences coded by nucleic acids originating from biological samples by searching a sequence database comprising nucleic acid coding sequences homologous to sequences encoding FP1 and FP2, retrieving sequence-identifying information for each homologous nucleic acid, choosing 1 of the retrieved sequences as the first gene to represent the origin of FP1 or FP2 and updating a computer-implemented data-store with the information that the first and second genes code for FP1 and FP2 and that a pair-wise interaction between the proteins evidences PPI;

(8) a computer-readable medium comprising instructions for causing at least 1 computers to function; and

(9) a computer system for storing and processing data related to pair-wise interaction between protein sequences encoded by nucleic acid obtained from biological samples comprising at least 1 computer memory provided with data structures for information representing an identity of selected first and second genes, an indication that these genes code for proteins involved in PPI, an indication that evidences PPI, and the

locations of the genes.

USE - The **detection method** (D) is useful for identifying inhibitors of PPI for therapeutic use, and for **detecting** specific **cell** types, tissue types, stage of development and disease states.

ADVANTAGE - From the population of the proteins characteristic of the particular tissue or a **cell-type**, all possible **detectable** PPI that occur can be identified and genes encoding these proteins can be **isolated**. Thus, parallel **analysis** of two **cell** types enumerates PPI that are common to both and those that are specific to both. This **analysis** has significant value since PPI specific to a disease state can serve as therapeutic points of intervention. Inhibitors of PPI can also be **isolated** in **rapid** fashion. The number of false positives and low throughput are reduced.

DESCRIPTION OF DRAWING(S) - The diagram shows a yeast interaction matting assay for the **detection** of protein-protein interactions (PPI). X is a DNA binding domain fusion and Y is an activation domain fusion protein. The activation of the DNA-binding domains are indicated as A and D respectively.

Dwg.2/30

L25 ANSWER 24 OF 24 WPIDS COPYRIGHT 2003 THOMSON DERWENT on STN
 ACCESSION NUMBER: 1999-357620 [30] WPIDS
 CROSS REFERENCE: 2000-551637 [51]
 DOC. NO. NON-CPI: N1999-266248
 DOC. NO. CPI: C1999-105798
 TITLE: **Isolating** genes encoding proteins with selected properties, useful for identifying therapeutic agents or targets.
 DERWENT CLASS: B04 D16 S03
 INVENTOR(S): BAILEY, J E; KOLLER, D; ORBERGER, G H; RENNER, W A
 PATENT ASSIGNEE(S): (CYTO-N) CYTOS BIOTECHNOLOGY GMBH; (CYTO-N) CYTOS BIOTECHNOLOGY AG
 COUNTRY COUNT: 84
 PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 9925876	A1	19990527 (199930)*	EN	136	
RW:	AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL OA PT SD SE SZ UG ZW				
W:	AL AM AT AU AZ BA BB BG BR BY CA CH CN CU CZ DE DK EE ES FI GB GD GE GH GM HR HU ID IL IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT UA UG US UZ VN YU ZW				
AU 9914155	A	19990607 (199943)			
NO 9904220	A	19990927 (200002)			
EP 972086	A1	20000119 (200009)	EN		
R:	AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU MC NL PT SE SI				
US 6197502	B1	20010306 (200115)			
NZ 337297	A	20010330 (200121)			
AU 760584	B	20030515 (200337)			

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 9925876	A1	WO 1998-US24520	19981117
AU 9914155	A	AU 1999-14155	19981117

NO 9904220	A	WO 1998-US24520	19981117
		NO 1999-4220	19990831
EP 972086	A1	EP 1998-958036	19981117
		WO 1998-US24520	19981117
US 6197502	B1	US 1997-972218	19971117
NZ 337297	A	NZ 1998-337297	19981117
		WO 1998-US24520	19981117
AU 760584	B	AU 1999-14155	19981117

FILING DETAILS:

PATENT NO	KIND	PATENT NO
AU 9914155	A Based on	WO 9925876
EP 972086	A1 Based on	WO 9925876
NZ 337297	A Based on	WO 9925876
AU 760584	B Previous Publ. Based on	AU 9914155 WO 9925876

PRIORITY APPLN. INFO: US 1997-972218 19971117

AN 1999-357620 [30] WPIDS

CR 2000-551637 [51]

AB WO 9925876 A UPAB: 20030612

NOVELTY - Identification of a recombinant nucleic acid (I) encoding an exogenous protein (II) having a selected property comprises preparing a population of eukaryotic host **cells**, culturing the **cells** under suitable conditions and identifying **cells** that **contain** (I).

DETAILED DESCRIPTION - Identification of a recombinant nucleic acid (I) encoding an exogenous protein (II) having a selected property comprises:

(a) preparing a population of eukaryotic host **cells**, each **containing** an expression system that includes a different recombinant nucleic acid (A), encoding an exogenous protein (B), linked to a control element;

(b) culturing the **cells** under conditions where (B) is expressed but expression of endogenous proteins is suppressed; and

(c) identifying **cells** that **contain** (I).

Alternatively, the expression system is a recombinant virus **containing** (A).

INDEPENDENT CLAIMS are also included for the following:

(1) generation of a genetic expression library encoding proteins having a predetermined property of interest comprises:

(a) providing a **composition** comprising a plurality of eukaryotic host **cells**, each **containing** an expression system that includes a different (A), encoding (B), linked to a control element;

(b) culturing the **cells** under conditions where (B) is expressed but expression of endogenous proteins is suppressed; and

(c) identifying **cells** that **contain** (I).

(2) genetic expression library comprising expression systems (or recombinant viruses) encoding (II) with a predetermined **cellular** localization, produced by (1);

(3) library of (II) produced by (1);

(4) nucleic acid library comprising a population of eukaryotic expression systems in different members, i.e. expression systems or recombinant viruses, having (I) linked to a control element.

USE - The **method** is used to sort genes according to the type of proteins they express, and also to identify new ligand/receptor interactions.

Typical applications of (I) and (II) are in **isolation** of new growth factors, cytokines, membrane receptors, cytoplasmic, organelle or nuclear proteins, all of which may be useful as therapeutic agents or therapeutic targets, e.g. apoptosis-promoting or tumor suppressing proteins, regulators of **cell** proliferation or metabolic processes etc.

(II) can also be used to screen for specific modulators.

Antibodies raised against (II) can be used for studying expression, function etc. of (II), also as diagnostic or therapeutic agents (optionally in the form of immunotoxins).

(I) can also be used as sources of therapeutic antisense or ribozyme sequences.

ADVANTAGE - The **method** allows the protein (rather than a partial DNA sequence) to be **isolated** and, since a wide range of **cells** can be used, they can be expressed with the correct glycosylation pattern.

(I) can be identified, characterized and **isolated** **quickly**; full-length cDNA is obtained in a single cloning step and proteins may be purified **quickly** without detailed knowledge of their properties. In situ labeling of (II) can be done, providing **rapid** identification of binding sites, and the **method** can produce vectors (for gene transfer) directly.

Dwg.0/15

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FILE 'HCAPLUS' ENTERED AT 15:48:27 ON 22 DEC 2003

L5 14820 SEA ABB=ON (?PROTEIN? OR ?PEPTIDE?) (W) (?MOLECUL? OR ?POPUL?)
 L6 3634 SEA ABB=ON L5 AND (?ISOLAT? OR ?DETECT? OR ?EXTRACT?)
 L7 1486 SEA ABB=ON L6 AND (?APPARATUS? OR ?MECHANISM? OR ?EQUIP? OR
 ?CONTAIN?)
 L8 671 SEA ABB=ON L7 AND (?PRODUCT? OR ?COMPOS? OR ?METHOD? OR KIT?)

FILE 'HCAPLUS' ENTERED AT 15:58:51 ON 22 DEC 2003

L9 300 SEA ABB=ON L8 AND ?ANAL?
 L10 144 SEA ABB=ON L9 AND (?LYSIS? OR ?FILTR?)
 L11 38 SEA ABB=ON L10 AND ?CHROMATOG?
 L12 3 SEA ABB=ON L11 AND ?RAPID?
 L13 4358 SEA ABB=ON L5 AND ?CELL?
 L14 25934 SEA ABB=ON (?PROTEIN? OR ?PEPTIDE?) (W) (?MOLECUL? OR ?POPUL?
 OR ?CELL?)
 L15 5686 SEA ABB=ON L14 AND (?ISOLAT? OR ?DETECT?)
 L16 2254 SEA ABB=ON L15 AND (?APPARATUS? OR ?MECHANISM? OR ?EQUIP? OR
 ?CONTAIN?)
 L17 979 SEA ABB=ON L16 AND (?PRODUCT? OR ?COMPOS? OR ?METHOD? OR
 KIT?)
 L18 303 SEA ABB=ON L17 AND (?LYSIS? OR ?FILT?)
 L19 167 SEA ABB=ON L18 AND ?CELL?
 L20 4 SEA ABB=ON L19 AND ?PORE?
 L21 42 SEA ABB=ON L19 AND ?CHROMATOG?
 L22 46 SEA ABB=ON L20 OR L21 *46 cits from CA Place*

FILE 'MEDLINE, BIOSIS, EMBASE, WPIDS, JICST-EPLUS, JAPIO' ENTERED AT
 16:14:04 ON 22 DEC 2003

L23 221 SEA ABB=ON L22
 L24 200 DUP REMOV L23 (21 DUPLICATES REMOVED)
 L25 24 SEA ABB=ON L24 AND (?RAPID? OR ?QUICK?)

*24 cits from
Other databases*

Inventor Search

Mohamed 09/903,864

22/12/2003

=> d ibib abs ind 14 1-1

L4 ANSWER 1 OF 1 HCPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 2002:72260 HCPLUS
DOCUMENT NUMBER: 136:115101
TITLE: Methods and compositions for rapid protein and peptide extraction and isolation from cells using a pore-containing lysis matrix
INVENTOR(S): Blakesley, Robert W.; Flynn, Barbara; Clausen, Peter
PATENT ASSIGNEE(S): Invitrogen Corporation, USA
SOURCE: PCT Int. Appl., 83 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002006456	A1	20020124	WO 2001-US22080	20010713
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
US 2002012982	A1	20020131	US 2001-903864	20010713
EP 1301591	A1	20030416	EP 2001-952705	20010713
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
PRIORITY APPLN. INFO.:			US 2000-218081P	P 20000713
			US 2001-274630P	P 20010312
			WO 2001-US22080	W 20010713

AB The present invention relates generally to compns., methods and kits for use in extracting and isolating protein or peptide mols. More specifically, the invention relates to such compns., methods and kits that are useful in the isolation of protein or peptide mols. from cells (e.g., bacterial cells, animal cells, fungal cells, viruses, yeast cells or plant cells) via lysis and one or more addnl. isolation procedures, such as one or more filtration and/or chromatog. procedures. In particular, the invention relates to compns., methods and kits wherein protein or peptide mols. are isolated using an integrated pore-containing lysis/filtration matrix, which may comprise one or more supports (e.g., polyolefin, sintered polyethylene, nitrocellulose, polypropylene, polycarbonate, cellulose acetate, silica, and the like). The compns., methods and kits of the invention are suitable for isolating a variety of forms of protein or peptide mols. from cells. The compns., methods and kits of the invention are particularly well-suited for rapid isolation of recombinant protein or peptide mols. expressed in bacterial cells, either as soluble protein, or as an inclusion body. The invention is particularly useful in high throughput applications, allowing quick isolation and/or anal. of proteins and/or peptides from numerous sources.

IC ICM C12N009-00

ICS C12N001-06

CC 9-9 (Biochemical Methods)

ST lysis filtration matrix protein peptide extn isolation

IT Purification
(affinity; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Filters
(bags, extraction apparatus housing; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Membrane, biological
(bilayer, disruption of; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Containers
(boxes, extraction apparatus housing; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Containers
(cartons, extraction apparatus housing; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Filters
(cartridges; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Denaturants
(chaotropic; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Containers
(extraction apparatus containing; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Ampuls
Bags
Bottles
Extraction columns
Pipes and Tubes
Vials
(extraction apparatus housing; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Buffers
Detergents
(lysis/disruption/permeabilization composition containing; methods and compns.
for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Enzymes, analysis
RL: ARG (Analytical reagent use); ARU (Analytical role, unclassified); NUU (Other use, unclassified); ANST (Analytical study); USES (Uses)
(lysis/disruption/permeabilization composition containing; methods and compns.
for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Acids, analysis
Bases, analysis
RL: ARU (Analytical role, unclassified); NUU (Other use, unclassified); ANST (Analytical study); USES (Uses)
(lysis/disruption/permeabilization composition containing; methods and compns.

for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Permeability
(lysis/disruption/permeabilization composition; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Polyamides, analysis
Polyesters, analysis
Polyolefins
Polysaccharides, analysis
RL: ARU (Analytical role, unclassified); NUU (Other use, unclassified); ANST (Analytical study); USES (Uses)
(matrix; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Animal cell
Bacteria (Eubacteria)
Cell
Chromatographic stationary phases
Cytolysis
Escherichia coli
Extraction
Extraction apparatus
Filters
Filtration
Frits
Fungi
Inclusion bodies
Plant cell
Pore size
Porous materials
Saccharomyces
Solubilization
Sonication
Yeast
(methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Peptides, analysis
RL: ANT (Analyte); PUR (Purification or recovery); ANST (Analytical study); PREP (Preparation)
(methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Antibodies
Coenzymes
Ligands
Nucleic acids
RL: ARG (Analytical reagent use); ARU (Analytical role, unclassified); NUU (Other use, unclassified); ANST (Analytical study); USES (Uses)
(methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Polymers, analysis
Salts, analysis
RL: ARU (Analytical role, unclassified); NUU (Other use, unclassified); ANST (Analytical study); USES (Uses)
(methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Plates
(multi-well, extraction apparatus housing; methods and compns. for rapid protein
and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Ceramics
(porous, matrix; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Proteins
RL: ANT (Analyte); NUU (Other use, unclassified); PUR (Purification or recovery); ANST (Analytical study); PREP (Preparation); USES (Uses)
(recombinant; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT Proteins
RL: ANT (Analyte); PUR (Purification or recovery); ANST (Analytical study); PREP (Preparation)
(separation; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

and

IT Infection
(viral, cell infected by virus; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT 57-13-6, Urea, analysis
RL: ARU (Analytical role, unclassified); NUU (Other use, unclassified); ANST (Analytical study); USES (Uses)
(chaotropic agent; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT 9001-63-2, Lysozyme 9011-93-2, Lysostaphin 9025-37-0, Zymolase
RL: ARU (Analytical role, unclassified); NUU (Other use, unclassified); ANST (Analytical study); USES (Uses)
(lysis/disruption/permeabilization composition containing; methods and compns.
for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT 7631-86-9, Silica, analysis 9004-34-6, Cellulose, analysis 9004-35-7,
Cellulose acetate 9004-70-0, Nitrocellulose
RL: ARU (Analytical role, unclassified); NUU (Other use, unclassified); ANST (Analytical study); USES (Uses)
(matrix; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

IT 9002-88-4, Polyethylene
RL: ARU (Analytical role, unclassified); NUU (Other use, unclassified); ANST (Analytical study); USES (Uses)
(sintered, matrix; methods and compns. for rapid protein and peptide extraction and isolation from cells using pore-containing lysis matrix)

REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT